DGT thresholds adaptation
Cobalt – copper – manganese – zinc

Report/Deliverable by

Isabelle Amouroux, Ifremer
Stéphane Guesdon, Ifremer
Jean-Louis Gonzalez, Ifremer
Florence Menet, Ifremer
Mélissa Dallet, Ineris

Table of Contents

Introduction

In the framework of the WFD (Water Framework Directive) (Directive 2000/60/EC)(1), each Member State (MS) has the ability to designate some pollutants as relevant at a hydrographic bassin or national stage. For these pollutants named River Basin-Specific Pollutants (RBSP), Environmental Quality Standards (EQS) are established by each Member Sate. These EQS should be determined on the basis of the methodology established at a European scale (TGD-EQS) (2) as described in the WP6 - action 1 report, but there is one difference. Instead of determining Quality standard for the 4 compartments: water column ($QS_{water\ eco}$), sediment ($QS_{sediment}$), biota ($QS_{biota\ sec\ pois}$), human health, through secondary poisoning ($QS_{biota\ hh}$) and through water consumption ($QS_{dw\ hh}$), they should be set for 3: water, sediment and biota as the protection goals are specifically ecological and do not cover the human health.

Additionnally to the priority substances (Cd, Ni, Pb), in the MONITOOL project, the DGT - Chelex (C-LSNM) was used for measuring the concentrations in marine water of 5 metallic compounds: cobalt, copper, iron, zinc and manganese. Other metals can be measured with this chelex resin: aluminium, chromium and silver, but as their results were considered as informative (http://www.dgtresearch.com/), they will not be contemplated in the rest of the document.

This document focuses on five metallic compounds, that can be identified as specific pollutants by MS: cobalt, copper, iron, zinc and manganese.

Scope

As DGT results cannot be directly compared to marine water thresholds, the specific objectives of this document is i) to propose a methodology so that DGT results can be applied in a regulatory context notably for establishing the ecological status of waterbodies as regard specific pollutants and ii) to adapt existing thresholds, defined in marine water at national level by a Member State, into DGT thresholds, for cobalt, copper, iron, zinc and manganese.

In order to reach these objectives, an overview of existing thresholds defined for Co, Cu, Fe, Zn and Mn was done, based both on a threshold research in existing databases and on a survey carried on among the MONITOOL partners on the specific pollutant they have defined.

The methodology defined for Pb, Cd, and Ni (WP6 - action 1) is declined for Co, Cu, Fe, Zn and Mn.
Based on the robust MONITOOL database of dissolved and labile metal concentrations in transitional and coastal waters, a data processing was done in order to propose adapted DGT thresholds.

1. Specific pollutants and existing thresholds

According to the WFD, for Specific Pollutants (Annex VIII substances), each Member State shall establish their EQSs (according to Annex V, Section 1.2.6 of the WFD) (1). In deriving environmental quality standards for pollutants (listed in points 1 to 9 of Annex VIII) for the protection of aquatic biota, Member States shall act in accordance with the following provisions. Standards may be set for water, sediment or biota.

Two complementary actions were done for the research of Co, Cu, Fe, Zn and Mn thresholds: a survey to know if these metals were defined as specific pollutants among the countries involved in MONITOOL, and a research on existing thresholds (EQS, QS, PNEC marine water) in databases.

1.1. Survey among MONITOOL partners

A survey was carried out among MONITOOL partners to get an overview of the metals that are defined as specific pollutant in their country. If the MONITOOL partner was not involved in the WFD monitoring or chemical and ecological assessment of waterbodies, this request was sent to the competent body so that it could be fullfiled.

The main questions of this survey were: are these metals (Co, Fe, Cu, Zn, Mn) listed as specific pollutants for marine water in your country? If so, is there thresholds defined for these metals in marine water or other matrixes? And what methodology and ecotoxicological data were used to derive these thresholds?

Answers were collected for UK, Irland, Scottland, France, Spain, Portugal and Italy.

Among these countries, only few listed at least one of these compounds as specific pollutant in marine waters. In total four compounds were listed:

- Copper and zinc in Spain,
- Copper, zinc, iron and manganese in England (UK).

It can be noticed that chromium is listed as Specific Pollutant in three countries: England, Italy and Spain.

Thresholds that are defined at national level, for marine water, are precised in Table 1.

Table 1: Specific Pollutants: thresholds in marine water defined by member states for Cu, Zn, Fe and Mn

	GB, England ($\mu g.L^{-1}$)	Spain ($\mu g.L^{-1}$)*
Copper (Co)	3.8	25
Zinc (Zn)	6.8	60
Iron (Fe)	1,000	-
Manganese (Mn)	123	-

** Annex V of Royal Decree 817/2015

It should be underlined that no information was given on how these thresholds were calculated, there is no information about the ecotoxicological data used for their determination.

This survey was completed following a research of existing thresholds on those metals.

1.2. Existing thresholds in referenced databases

For this project, a search of existing thresholds in different online databases was conducted. These databases are:

- European Chemicals Agency (ECHA) website (https://echa.europa.eu/fr/home). ECHA is an European Union agency that engages the regulatory authorities for the implementation of EU chemicals legislation such as the REACH Regulation (Registration, Evaluation, Authorisation and Restriction of Chemicals),
- ETOX (Information System Ecotoxicology and Environmental Quality Targets, https://webetox.uba.de/webETOX/index.do), is a German database managed by UBA (German Environment Agency) allowing access to ecotoxicity data for aquatic and terrestrial organisms. This database contains information on various national and international environmental quality guidelines, targets, standards, criteria, and limit values. In particular for substances which are relevant for development of environmental quality standards according to the Water Framework Directive 2000/60/EC.
- Chemical Substance Portal (CSP, https://substances.ineris.fr/fr/), a French database managed by INERIS (French National Institute for Industrial Environment and Risks) allowing access on toxicology, ecotoxicology and technical-economics data.

Results of this search are presented in Table 2. When several thresholds were found for the same substance, we selected the one that appears to be the most relevant, based on expert judgment and some criteria. Given the aims of the MONITOOL project, the national **regulatory thresholds** proposed within the WFD framework for "River Bassin Specific Pollutant (RBSP)" were prioritized. Then, it is important to look at the "**context of determination of the thresholds found".** Threshold reviewed among European working groups are considered robust (for instance, thresholds (PNEC) proposed within the Risk Assessment Report (RAR) carried out in the context of the older Council Regulation (EEC) No. 793/93 on the evaluation and control of existing substances) or within the evaluations carried out under REACH. Contrarily, PNECs defined by manufacturers within the REACH regulation (registration dossier) must be considered carefully as, most of them, have not been subjected to any evaluation. The **ecotoxicity dataset** used for threshold derivation can also give indications on its robustness.

Table 2: Existing thresholds for cobalt, copper, iron, manganese and zinc

Substance / Cas Num	Available thresholds	Source	Details on thresholds derivation
Cobalt / 7440-48-4	$PNEC_{freshwater}$ = 0.5 µg/L (Assessment factor methodology, AF = 10) $PNEC_{marine\ water}$ = 0.1 µg/L (Assessment factor methodology, AF = 50)	PCS (Ineris datasheet, 2006 – In French)	Only two thresholds found for marine water. The first derived by Ineris (French institut) in 2006 and the second derived by manufacturer within REACH regulation (registration file). INERIS, 2006: Few available marine water organisms ecotoxicity data (only 3 results: 2 for crustaceans and 1 for fish). The $PNEC_{marine\ water}$ was determined from freshwater ecotoxicity test result (*Daphnia magna*, NOEC (21j) = 5 µg/L) with an AF of 50.
	$PNEC_{freshwater}$ = 0.62 µg/L (SSD methodology, AF = 3) $PNEC_{marine\ water}$ = 2.36 µg/L (SSD methodology, AF = 3) $PNEC_{sediment\ freshwater}$ = 53.8 mg/kg sed dw (Assessment factor methodology, AF = 10) $PNEC_{sediment\ marine\ water}$ = 69.8 mg/kg sed dw (Assessment factor methodology, AF = 10)	ECHA Website (registration file)	ECHA Website (registration file): It is very difficult to evaluate the robustness of the PNEC proposed within the Registered substances factsheets (ECHA website) because few details about the ecotoxicity dataset used and methodology applied are available in these files. Nevertheless, in this case, it is mentioned that the SSD methodology is applied which implies the use of an important ecotoxicity data set.
	Suizerland (1994): QZ (Quality Standard, protection of aquatic life, general) = 50 µg/L (total dissolved) Netherland (1999): BACKG (background concentration, Quality objective, surface water) = 0,2 µg/L (dissolved). Germany (2003): Quality Criteria, aquatic life, general (QN-V) = 0.9 µg/L (SSD metgodology, AF = 5), PNEC = 0.9 + BACKG Netherland (2005). MPA (Maximum Permissible Addition, Quality criteria, aquatic life protection, freshwater) = 0.5 µg/L (dissolved); MPC (Maximum Permissible Concentration, surface water, = 0.69 µg/L (dissolved)	ETOX database (only freshwater thresholds)	Conclusion: For the reasons mentioned above, neither of the two available thresholds can be considered robust. In this case the most protective threshold should be used.
Copper / 7440-50-8	$PNEC_{freshwater}$ = 1.6 µg/L (SSD methodology, AF = 2) $PNEC_{marine\ water}$ = 0.8 µg/L (SSD methodology, AF = 2) $PNEC_{sediment\ freshwater}$ = 0.8 mg/kg dw (Assessment factor methodology, AF = 50) $PNEC_{secondary\ poisonning}$ = 5.7 mg Cu/kg food (AF = 30)	PCS (Ineris datasheet, 2004 – In French)	INERIS, 2004: Fresh and saltwater data are pooled (same sensitivity). 80 differents species. SSD methodology applied: HC5 = 1,6 µg/L, AF = 2. $PNEC_{marine\ water}$ = 0.8 µg/L ECHA Website: It is very difficult to evaluate the robustness of the PNEC proposed within the Registered substances factsheets (ECHA website) because few details about the ecotoxicity dataset used and methodology applied are available in these files. Nevertheless, in this case, it is mentioned that the SSD methodology is applied which implies the use of an important ecotoxicity data set.
	$PNEC_{freshwater}$ = 7.8 µg/L (SSD methodology, AF = 1) $PNEC_{marine\ water}$ = 5.2 µg/L (SSD methodology, AF = 1) (DOC normalised to 2 mg/L) $PNEC_{sediment\ freshwater}$ = 87 mg/kg sed dw (Equilibrium partitioning method)	ECHA Website (registration file)	

	$PNEC_{\text{sediment marine water}}$ = 676 mg/kg sed dw (Equilibrium partitioning method)		**Proposed EQS - United Kingdom (UK TAG, 2011):** Increasing dissolved organic carbon (DOC) has been shown to significantly reduce the ecotoxicity of Cu in freshwater and marine water. Each individual NOEC/L(E)C10 value (data from Voluntary Risk Assessment of Copper (ECI, 2008) and a further literature search for saltwater chronic data) was normalised using the DOC correction. 29 species NOECs used, an HC5 = 2.64 µg Cu/L was generated (SSD methodology, AF = 1). $PNEC_{\text{reference saltwater}}$ = 2.64 µg/L (DOC normalised to 1 mg/L). This PNEC is then adjusted to ambient conditions through the use of a bioavailability correction based on the concentration of DOC. Site specific $PNEC_{\text{saltwater}}$ = 2.64 + (2.677 x ((0.5 x DOC) – 0.5)) (when an AF of 1 is applied in deriving the reference PNEC). **EQS datasheet - Germany in 2015:** The original non-normalised data presented in the UK-TAG assessment (2011) were used. For the SSD, 38 marine species belonging to eight taxonomic groups (fish (n = 2), crustaceans (n = 12), mollusca (n = 10), polychaeta (n = 2), echinoderma (n = 3), coelenterate (n = 1), cnidarians (n = 3), algae (n = 5)) were considered. HC5 = 4.69 µg/L. The approach for HC5 derivation using non normalised toxicity data shows a high intraspecies variability in NOEC values for some species. Due to the partly high intraspecies variability of the NOEC values and since the data cover the minimum number of taxonomic groups (n = 8), an assessment factor of 5 is proposed for the derivation of the $AA\text{-}EQS_{\text{generic, saltwater, eco}}$ of 0.7 µg Cu/L in terms of non-normalised total copper concentration. Conclusion: It is proposed to retain the most recent marine threshold (EQS) proposed by Germany.
	Canada (1987): Canadian water quality guidelines, protection of aquatic life, freshwater, 2-4 µg/L (Total) Switzerland (1994). Quality Standard (QZ), protection of general aquatic life = 10 µg/L (total dissolved) Netherland (1999): Maximum Permisible Concentration (MPC), surface water protection = 1.5 µg/L (dissolved) and BACKG (background concentration, Quality objective, surface water) = 0.4 µg/L (dissolved) USA (1999): National recommended water quality criteria-correction (CCC), protection of aquatic life, salt water = 3,1 µg/L (dissolved) United Kingdom (UK TAG, 2011): Proposed EQS for Water Framework Directive Annex VIII substances: copper (saltwater) (for consultation). $PNEC_{\text{reference saltwater}}$ = 2.64 µg/L dissolved Cu (SSD methodology, AF = 1) (DOC normalised to 1 mg/L). $PNEC_{\text{site specific saltwater}}$ = 2.64 + (2.677 x ((0.5 x DOC) – 0.5)) (if an assessment factor of 1 is to be applied in deriving the reference PNEC) Germany (2015): EQS datasheet. AA-QS (protection of aquatic life, salt water) = 0.7 µg/L (dissolved, SSD methodology with HC5 = 3.5 µg/L and AF = 5). AA-QS (protection of aquatic life, freshwater) = 1.1 µg/L (dissolved, SSD methodology with HC5 = 5.7 µg/L and AF = 5); Ireland: Target value = 0.5 µg/L (surface water, no further data)	ETOX database	
Iron / 7439-89-6	Canadian (1987): Canadian water quality guidelines (GL), protection of aquatic life freshwater = 300 µg/L (Total). United Kigdom (1994): Enviromental quality standards for United Kingdom Waters. Quality Standard (EQS; AA-QS) for freshwater and marine water = 1000 µg/L (dissolved).	ETOX database https://www.wfduk.org/sites/default/files/Media/iron.pdf	**Proposed EQS - United Kingdom (UK TAG, 2007):** Data for acute toxicity to marine species are available for five taxonomic groups (crustaceans, molluscs, annelids, echinoderms and fish). Chronic toxicity data are limited to algae and crustaceans. No chronic data found for fish. The saltwater data are based on nominal concentrations of iron and are, therefore, not suitable for PNEC derivation. The $PNEC_{\text{add*, saltwater}}$ is based on the lowest reliable long-term freshwater data (*Daphnia magna*, reproduction, NOEC (21d) = 0.16 mg/L) with an AF = 100. $PNEC_{\text{add, saltwater}}$ = 1.6 µg/L iron (dissolved). This PNEC is 625 times lower than the existing EQS of 1000 µg/L proposed

	USA (1999): National recommended water quality criteria-correction (CCC). Protection of aquatic life, freshwater = 1000 µg/L (Total, dissolved: not mentionned).		in 1994) (new data and differente derivation approach). In the proposed EQS facsheet, authors inform that current analytical methods may not be adequate to analyse iron for compliance with the proposed PNECs. The proposed PNECs are likely to be insignificant compared with backgrounds, in which case it may be appropriate to consider adopting the background. High uncertainty in the extrapolations to derive saltwater PNECs may be reduced by the generation of additional ecotoxicological data".
	United Kingdom (UK TAG, 2007). Proposed EQS for Water Framework Directive Annex VIII substances: Iron (total disolved). $PNEC_{add*,freshwater}$ = 16 µg/L. $PNEC_{add*,saltwater}$ = 1.6 µg/L.	UK TAG (2007) https://www.wfduk.org/sites/default/files/Media/iron.pdf	Australian default guideline values (DGV) (2020): Threshold based on total iron that includes dissolved, colloidal and precipitated forms of iron. Chronic toxicity data for iron were available for five species found in Australian and New Zealand marine water (Mussel *Mytilus galloprovincialis*, the oyster *Saccostrea glomerata*, the abalone *Haliotis rubra* and the corals *Acropora spathulata* and *Platygyra daedalea)*. Additional chronic data were reported from international waters for the crab Cancer *anthonyi* and a microalga *Isochrysis galbana*. Oyster (*S. glomerata*) larval development had the most sensitive endpoint, with a NOEC (48h) of 122 µg/L.
	Australia (2020). Toxicant default guideline values (DGV)**. Total iron in marine water, moderate reliability (SSD, Level of species protection 95%) = 180 µg/L.	https://www.waterquality.gov.au/ (Technical brief June 2020)	In the factsheet, authors inform that "It is unclear what forms of iron in marine water are responsible for toxicity to aquatic biota. Mechanisms such as iron-specific membrane-transport proteins and the production of ironbinding organic ligands (siderophores) are likely to be adversely affected when iron concentrations exceed requirements". Authors consider this threshold with "moderate reliability": based on the outcomes for the following three criteria: Sample siz = 9 (good); Type of toxicity data—two chronic EC10 values, five chronic NOEC values, two converted acute values; SSD model fit (Good)". Conclusion: For the reasons mentioned above, neither of the three available marine thresholds can be considered robust. In this case the most protective threshold should be used.
Manganese / 7439-96-5	$PNEC_{freshwater}$ = 15 µg/L (Assessment factor methodology, AF = 100) $PNEC_{marine\ water}$ = 1.5 µg/L (Assessment factor methodology, AF = 1000)	PCS (Ineris datasheet, 2003 – In French)	INERIS, 2003: Only one acute ecotoxicity data available for marine organism *Dytilum brightwellii*. For freshwater, only two chronic ecotoxicity data available for crustaceans and fish (not for algae witch seems to be the most sensible organisms based on acute ecotoxicity data). The $PNEC_{marine\ water}$ was derived from acute marine ecotoxicity test result (*Dytilum brightwellii*, EC50 = 1,5 mg/L) with an AF of 1000.
	$PNEC_{freshwater}$ = 34 µg/L (Assessment factor methodology, AF = 50) $PNEC_{marine\ water}$ = 3 µg/L (Assessment factor methodology, AF = 500) $PNEC_{sediment\ freshwater}$ = 3.3 mg/kg sediment dw (Assessment factor methodology, AF = 500) $PNEC_{sediment\ marine\ water}$ = 0.34 mg/kg sediment dw (Assessment factor methodology, AF = 5000)	ECHA Website (registration file submitted by the industrial)	

	United Kigdom (UK TAG, 2010): Proposed EQS for Water Framework Directive Annex VIII substances: manganese (bioavailable) (for consultation). Quality Criteria for Aquatic life Marine water protection (proposed PNEC) = 0.05 μg/L (dissolved, AF methodology). Quality Criteria for Aquatic life Freshwater water protection (proposed PNEC) = 123 μg/L (bioavailable, SSD methodology, HC5 = 246 μg/L, AF = 2).	ETOX database	ECHA Website: It is very difficult to evaluate the robustness of the PNEC proposed within the Registered substances factsheets (ECHA website) because few details about the ecotoxicity dataset used and methodology applied are available in these files. Proposed EQS - United Kingdom (UK TAG, 2010): The effects database for marine species is considerably smaller than that for freshwater organisms. Long-term saltwater data are available only for algae, annelids, crustaceans and molluscs. No long-term manganese toxicity data could be found for saltwater fish. The most sensitive and reliable long-term toxicity values relate manganese exposure over 7–20 days to growth of Pacific oyster, *Crassostrea gigas*, and hatching of yellow crab, *Cancer anthonyi*, both resulting in a lowest observed effect concentration (LOEC) of 10 μg/L. An assessment factor of 2 is applied to extrapolate to the NOEC from the LOECs of 10 μg l/L and another factor of 100 is recommended to account for interspecies differences in sensitivity because there are no long-term NOECs for saltwater fish or algae. This results in a $PNEC_{saltwater}$ of 0.05 μg/L manganese (dissolved). In the factsheet, authors say "This value is very low and is an order of magnitude below the low end of the range of concentrations reported in seawater and is therefore not implementable as an EQS". This PNEC should be considered as a PNEC $_{add}$ manganese is a naturally occurring substance that organisms will have been exposed to over an evolutionary timescale. In this case, the PNEC applies only to the 'added' contribution over and above the background level. Conclusion: For the reasons mentioned above, neither of the three available marine thresholds can be considered robust. In this case the most protective threshold should be used.
Zinc / 7440-66-6	$PNEC_{freshwater}$ = 20.6 μg/L (SSD methodology, AF = 1) $PNEC_{marine\ water}$ = 6.1 μg/L (SSD methodology, AF = 1) $PNEC_{sediment\ freshwater}$ = 117.8 mg/kg sediment dw (Equilibrium partitioning methodology) $PNEC_{sediment\ marine\ water}$ = 56.5 mg/kg sediment dw (Equilibrium partitioning methodology)	ECHA Website	ECHA Website: It is very difficult to evaluate the robustness of the PNEC proposed within the Registered substances factsheets (ECHA website) because few details about the ecotoxicity dataset used and methodology applied are available in these files. EQS datasheet (Germany, 2014): Marine species are well-represented with chronic NOECs for saltwater species from six taxonomic groups: algae (unicellular and multicellular), annelids, coelenterates, crustaceans, echinoderms, and molluscs. SSD methodology applied (HC5 = 6.2, AF = 2), $PNEC_{add}$ = 3.0 μg/L. In the factsheet authors say the "the background concentration for the North Sea is estimated to be 1 μg/L and used for the AA-QSmarine water for zinc, resulting in an overall marine AA-QS value of 3.0 μg/L + 1 μg/L= 4.0 μg/L.
	Canadian (1987): Canadian water quality guidelines (GL), protection of aquatic life, freshwater = 30 μg/L (Total) Switzerland (1994): Quality Standard (QZ), protection of general aquatic life = 200 μg/L (total dissolved).	ETOX database	

	USA (1999): National recommended water quality criteria-correction (CCC). Protection of aquatic life, marine water = 81 µg/L (Total, dissolved: not mentionned). Protection of aquatic life, freshwater = 120 µg/L (Total, dissolved: not mentionned). European Union (2008). European Union Risk Assessment Report zinc metal CAS No: 7440-66-6 EINECS No: 231-175-3. $PNEC_{add\ Aquatic\ life,\ fresh\ water\ protection}$ = 7.8 µg/L (hardness > 24 mg CaCO3/L) and 3.1 µg/L (hardness < 24 mg CaCO3/L) (Zn bioavailable, SSD methodology with AF = 2). $PNEC_{marin\ water}$: Same PNEC as for freshwater. United Kigdom (UK TAG, 2010): Proposed EQS for Water Framework Directive Annex VIII substances: zinc (For consultation). $PNEC_{add\ freshwater}$ = 10.9 µg/L zinc (bioavailable, SSD methodology, HC5 = 10.9 µg/L, AF = 1). $PNEC_{add\ saltwater}$ = 3.4 µg/L zinc (disolved, SSD methodology, HC5 = 6.76 µg/L, AF = 2). Germany (2014): EQS datasheet. QS-SW (protection of aquatic life, saltwater) = 3 µg/L (dissolved, SSD methodology with, HC5 = 6.2, AF = 2). QS-FW (protection of aquatic life, freshwater) = 10.9 µg/L (bioavailable, SSD methodology with AF = 1); QS sediment freshwater = 49 mg/kg dw (AF methodology, AF 10). Ireland: Target value = 2.3 µg/L (surface water, no further data)	This threshold is very close to the $PNEC_{add,saltwater}$ proposed by UK TAG in 2010. Conclusion: It is proposed to retain the most recent marine threshold (EQS) proposed by Germany.

*The 'added risk' approach could be appropriate when setting EQSs for iron. This is because iron is a naturally occurring substance that organisms will have been exposed to over an evolutionary timescale. In this case, the PNEC applies only to the 'added' contribution over and above the background level.

** A guideline value recommended for generic application in the absence of a more specific guideline value (e.g. a site-specific guideline value), in the Australian and New Zealand Guidelines for Fresh and Marine Water Quality. Formerly known as "trigger values".

1.3. Thresholds considered for MONITOOL Project

The thresholds considered for MONITOOL project are presented in Table 3. They have been selected following a first level of expert judgment (the expertise of ecotoxicological data for the derivation of the threshold was not done in the context of this work). These thresholds are considered to be the most robust or the most recent among existing thresholds presented in Table 2, and in the case none of the available marine thresholds can be considered robust (cobalt, iron, manganese), then the most protective threshold should be used.

Table 3: Thresholds considered for MONITOOL project

Substance	Thresholds ($\mu g.L^{-1}$)
Cobalt (Co)	$PNEC_{\text{marine water}} = 0.1$ *(Ineris)*
Copper (Cu)	AA-QS (protection of aquatic life, salt water)= 0.7 *(EQS Datasheet, Germany 2015)*
Iron (Fe)	$PNEC_{\text{add marine water}} = 1.6$ *(UK TAG, 2007)*
Manganese (Mn)	$PNEC_{\text{add marine water}} = 0.05$ *(UK TAG, 2010 – proposed EQS)*
Zinc (Zn)	$PNEC_{\text{add marine water}} = 3$ *(Germany, 2014 – proposed EQS)*

Manganese, iron and zinc have a PNEC $_{\text{"add marine water"}}$. This 'added risk' approach could be appropriate when setting thresholds for some metals. This is because metals are naturally occurring substance that organisms will have been exposed to over an evolutionary timescale. **In this case, the PNEC applies only to the 'added' contribution over and above the background level.**

$$PNEC_{\text{marine water}} = PNEC_{\text{add marine water}} + \text{background concentration}$$

If the metal was listed as specific pollutant, thresholds defined at national level (§ 1.1.) were also considered for the adaptation of the DGT-threshold, for information purpose only (no comments will be done on these adaptations as no information are provided on how these thresholds were determined).

2. Determination of adapted thresholds in DGT

The adaptation of the threshold defined in marine water for a metal to Threshold $_{DGT}$ implies the establishment for each metal considered a valid relationship between the dissolved concentration measured in spot water samples and the DGT-labile concentration measured by DGTs (Figure 1). A relationship can be defined according to the following generic equation (equation 1):

$$[M]_{DGT} = slope \times [M]_{Dissolved\ fraction} + intercept \qquad (eq.1)$$

From equation 1, the Threshold $_{DGT}$ corresponding to the Threshold defined in marine water value is determined, based on metal concentrations measured by both techniques and using a predictive model.

DGT results can then be compared to this Threshold $_{DGT}$.
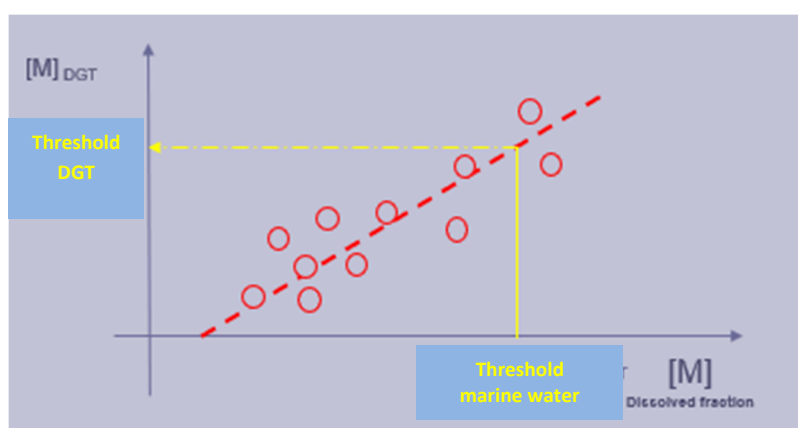
Figure 1: Illustration of the relationship between [M] in DGT and in the dissolved fraction, and determination of Threshold $_{DGT}$

$[M]_{DGT} = slope \times [M]_{Dissolved\ fraction} + intercept$

This requires having results, acquired in parallel, from spot water samples and DGTs. The data must be reliable, homogeneous, and with low analytical uncertainty at the required low concentration levels. The sites sampled should cover a concentration range as wide as possible (from slightly contaminated to highly contaminated sites). The data must be sufficiently numerous to allow obtaining a robust relationship.

The objective is to allow the establishment of relationships between the concentrations measured in the DGTs and the concentrations measured in the dissolved fraction (spot-sampling) by ICP-MS. For each sampling point and each season (wet season and dry season), a DGT result (average of the triplicates) and a spot-sampling result (average of the discrete samples results obtained during the DGT immersion time) were obtained. In total 36 sampling points were sampled, allowing 84 couples of data (DGT – spot sampling) for the two sampling periods.

The MONITOOL protocol is described in WP5 (not described here) (3).

The following data processing is based on the MONITOOL dataset provided by WP4 (v24). This dataset provides valid data results, after a validation process carried out by the expert laboratories in charge of the analysis. The

following study of determination of Threshold$_{DGT}$ in relation to Threshold $_{marine\ water}$ is complementary to the work done in WP4, dealing with the study of the relationships between the metal concentrations measured by the different methods (i.e., DGT-labile metal concentrations and concentrations measured by ICP-MS and voltammetry in spot water samples) and the influence of environmental parameters (temperature, salinity, pH, SPM, COD) on these relations.
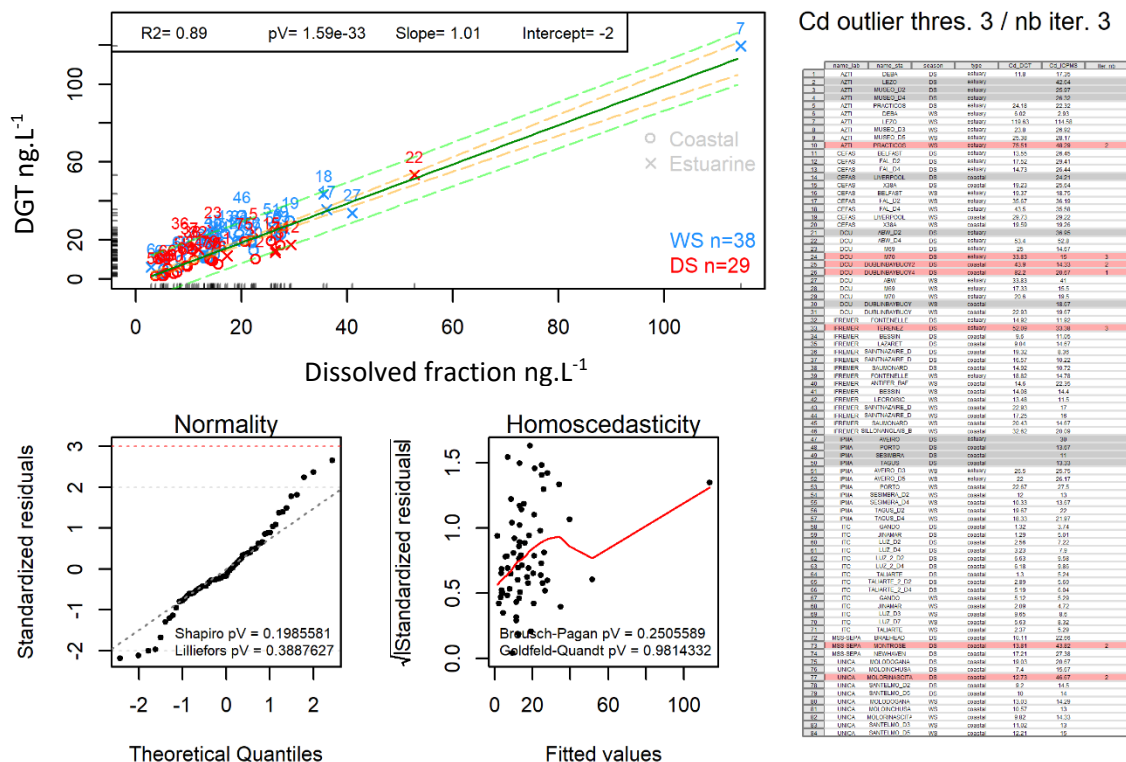
Data processing:

For each metal, a statistical process has been run to identify outliers based on the results of linear models. The objective here is to identify as outliers those samples presenting standardized residuals, from the linear model, greater than 3 (rejection of values above 99.73% of the total values assuming that their distribution follows a normal distribution).

In this way, an iterative process is carried out until no more outliers are identified: the process starts with the linear modelling of all the results, thus allowing the identification of possible outliers. A new linear model is made by removing the previously identified outliers. The new outliers are then removed and the process is repeated until all the outliers are identified. The last model without outliers is then kept and diagnosed.

The diagnosis of the model is based on a standard graphical panel with graphical and numerical outputs to judge the normality and homoscedasticity of the residuals. The description of the outputs is done Figure 2 for illustration purpose. Although there is a consensus on these parameters for the validation of linear models, the assumption of normality of the residuals is often considered secondary; the linear model is then considered to be robust to the absence of normality of the residuals given. Nevertheless, in addition to the QQ-plot (which allows a graphical analysis of the normality of the residuals) the Shapiro-Wilk and Lilliefors (Kolmogorov-Smirnov) tests were performed to guide the validation of the model (4). The most important thing is to have a QQ-plot curve that is as linear as possible with no real residuals that stand out (no curved distributed residuals). In this case, even if the results of the normality tests mentioned above are below the validity threshold (p-value < 0.05), the normality hypothesis is accepted (except when the p-value of the tests is far from the threshold).

The homoscedasticity of the residuals was also taken into account thanks to the graphical output illustrating the square roots of the residuals (standardized residuals) as a function of the predicted theoretical values (fitted-values) of Y and the Breush-Pagan and Golfeld-Quandt tests. Although we have taken care to use Koenker's version (less sensitive to the lack of normality than original version), the Breush-Pagan test stay sensitive to the lack of normality. The homoscedasticity of the residuals was also tested with the Golfeld-Quandt test.

The validity of the final model for each metal was evaluated, first based on its pValue (valid if <= 0.05), and secondly on the normality and homoscedasticity of the residues (valid if >= 0.05); It should nevertheless be specified that the choice to consider two tests for each criterion offers the possibility to be more objective on the decision (each test having its limits/gaps). Thus, the validation of a model is acquired if one of the 2 tests for each criterion is valid. As mentioned above, normality can be the subject of a less clear-cut decision, also based on the graphical output (QQ-plot). In any case, it is advisable to remain vigilant on this cut-off point of statistical thresholds (i.e. p<0.05), keeping in mind the number of values taken into account, their general appearance by a visual glance (graphical output) and by relativizing the concept of "significant" (5).

Figure 2: Illustration of standard output:

- Upper left chart shows scatterplot DGT=f(ICPMS)* with linear model based on white results illustrated in the table on the right; the dark green line corresponds to the linear model, which is surrounded by its confidence interval CI 95(orange dashed lines), and its predictive intervalle (PI 95) (green dashed lines). *or ICPMS=f(DGT)*;
- Bottom letf chart represents QQ-plot illustrating normality of residuals with Shapiro and Lilliefors tests (normality: pV > 0.05);
- Bottom right chart represents Residuals variance plot illustrating homogeneity of residuals with Breush-Pagan and Golfeld-Quandt tests (homogeneity: pV > 0.05);
- Table on the right specifies the identity of samples with DGT and ICPMS values ; background gray illustrate samples not considered because of one of DGT or ICPMS missing ; background red color identify samples which are outliers ; these two types of samples are not take into accompt in the regression. In some case, the number of iteration for outliers selection are specified.

When a model is valid:

- A concentration of metal in DGT can be predicted from its concentration measured in dissolved fraction,

$$[[M]_{DGT}= slope \times [M]_{Dissolved\ fraction} + intercept];$$

Based on this model the Threshold $_{DGT}$ (ex: PNEC $_{DGT}$ or AA-QS $_{DGT}$) can then be determined. Even if the model is valid, its use is reliable within its validity range. Ideally this Threshold $_{DGT}$ is determined in the validity range. If this is not the case, it could be determined in a pragmatic way as the projection of the Threshold $_{marine\ water}$ value, and in order to ensure that level of protection of the Threshold$_{DGT}$ is the same at least than the Threshold $_{marine\ water}$, it can be determined using the projection minus PI 95 (Prediction Interval 95%).

3. Determination of Threshold $_{DGT}$

3.1. Description of the MONITOOL dataset

The range of concentrations of Co, Cu, Mn and Zn obtained by the MONITOOL strategy and sampling sites are listed in Table 4. As iron results measured in dissolved fraction were not considered valid by expert laboratories, they are not presented in this document.

Table 4: Concentration of metals measured in dissolved fraction for Co, Cu, Mn and Zn.

Mean of concentration of metal in dissolved fraction (ICP-MS)	Mean ng.L^{-1}	Minimum ng.L^{-1}	Maximum ng.L^{-1}
Co	38	2	176
Cu	1,287	36	4,867
Mn	12,890	2,678	70,290
Zn	2,975	62	24,730

3.2. Cobalt

The graphical representation of the relationship between concentration of cobalt measured in dissolved fraction and the labile concentration measured in DGT is presented in Figure 3. The regression line is surrounded by its prediction interval (PI$_{95\%}$), the statistical process (Fig 3 a)) and the determination of Threshold $_{DGT}$ (Fig 3 b)) are presented. For cobalt, the model validation assumptions were partially satisfied but the relationship between DGT and dissolved concentrations measured by ICP-MS was significant. The model validation criteria were met (p-Values, homoscedasticity, …) with the exception of residue normality, which does not necessarily present a rebuttal aspect to the invalidation of the model, in addition the Lilliefors test p-value is not very far by the threshold of 0.05 (cf Data processing). The relationship between DGT and dissolved fraction was significant (Fig.4)), and can be described by the equation 3:

$$[Co]_{DGT} = 0.81 \, [Co]_{ICP-MS} + 81 \qquad \text{(Eq. 3)}$$
Concentrations are expressed in ng.L^{-1}

The only downside to this relationship is the coefficient of determination which indicates that only 23% of the variance of the measured concentrations in the DGTs is explained by the model. From this model, a concentration of metal in DGT can be predicted from its concentration measured in dissolved fraction. Based on the MONITOOL data, the upper limit of the validity area of this model is 175 ng.L^{-1}, the range of validity correspond to the area in light green (Figure 3 (b)). Beyond this limit, this model is not verified.
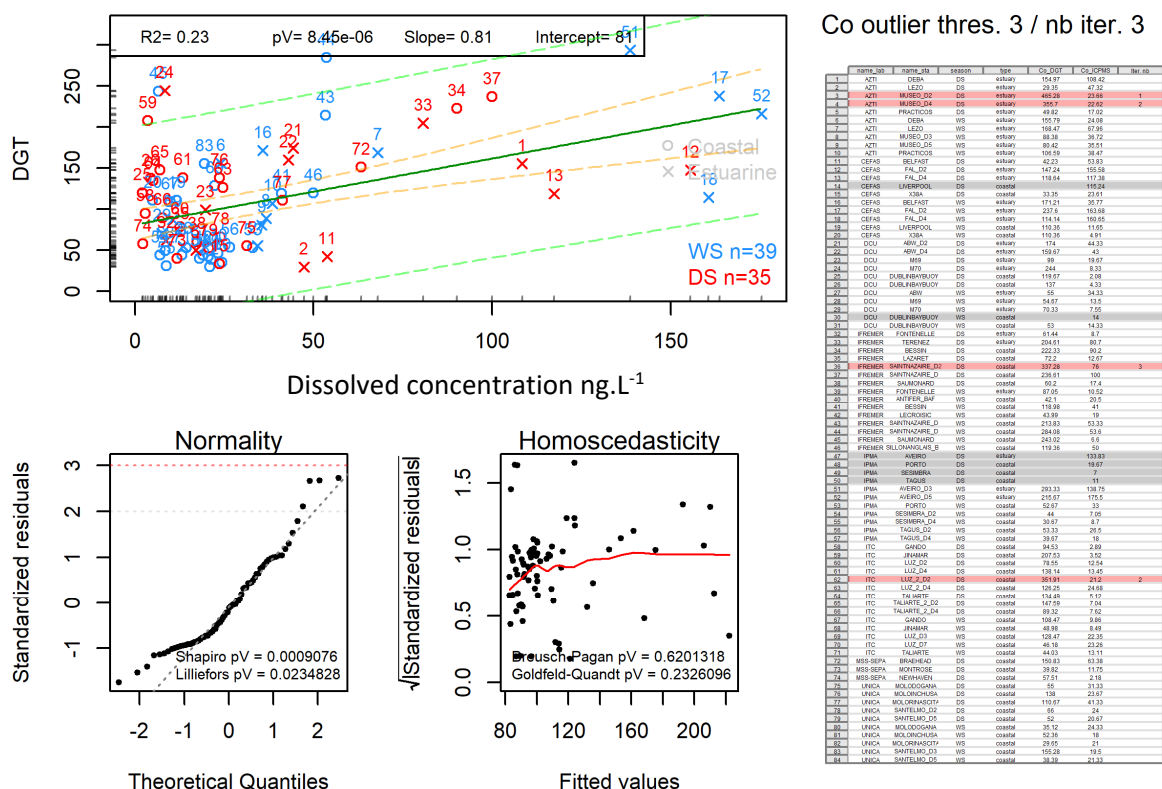
Figure 3 a): Cobalt: Relationship between dissolved fraction and DGT labile concentration - Statistical process. The dark green line corresponds to the linear model, which is surrounded by its confidence interval CI 95 (orange dashed lines), and its predictione interval (PI 95) (green dashed lines).
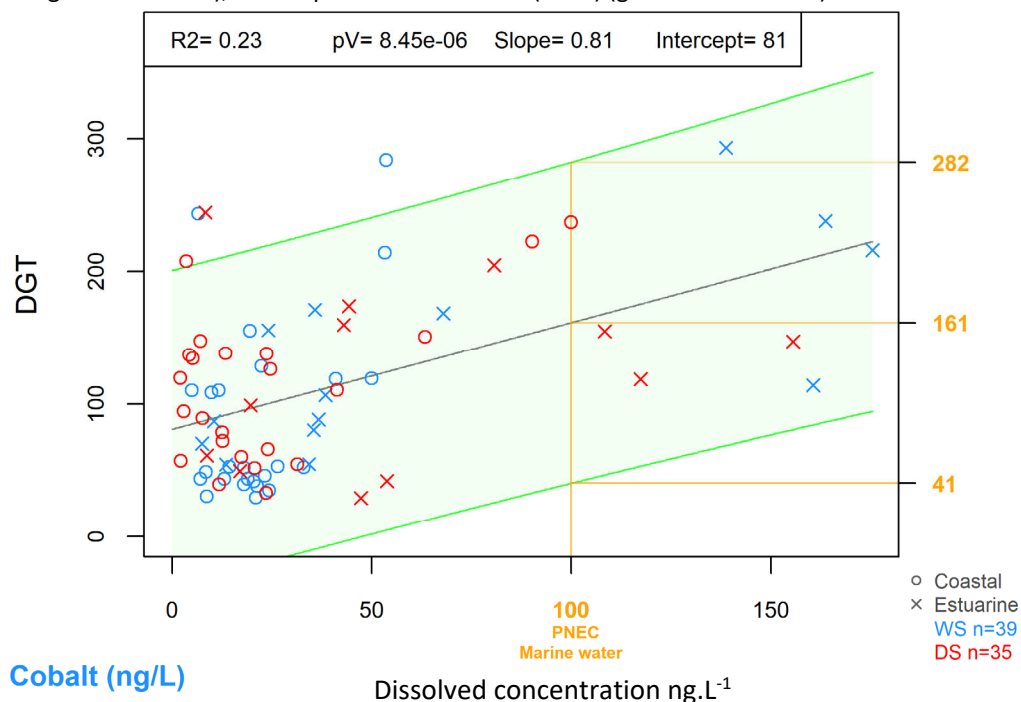
Figure 3 b): Cobalt: Relationship between dissolved fraction and DGT labile concentration - determination of one Threshold $_{DGT}$ – the regression line (black line) is surrounded by its prediction interval (PI 95) in green line in the range of validity (area in light green).

14

The threshold defined for cobalt (0.1 $\mu g.L^{-1}$, 100 $ng.L^{-1}$ in Figure 3b) is in the range of validity of the model. The predicted concentration in DGT for these thresholds is precised in table 5. In order to be more protective, the low value of the prediction interval can be selected as a proposed Threshold $_{DGT}$.

Table 5: Determination of Threshold $_{DGT}$ for cobalt

Threshold Marine water ($\mu g.L^{-1}$)	Threshold $_{DGT}$ n°1 - ($\mu g.L^{-1}$) Linear Model Regression	Threshold $_{DGT}$ n°2 - ($\mu g.L^{-1}$) Linear Model Regression minus low prediction interval (PI 95%)
0.1	0.16	0.04

As cobalt is not listed as specific pollutant in the member states involved in MONITOOL project, none other national threshold is defined.

An overview of the matching is done in Table 6 between assessment based on the dissolved fraction or DGT results. In the Table 7, results for each site and sampling campaign are compared to the thresholds: DGT results to Thresholds $_{DGT}$ n° 1 and 2 and dissolved concentration results to threshold marine water.

Table 6: Cobalt - Matching between assessment based on thresholds defined in marine water and in DGT

		[Co] dissolved fraction < 0.1 $\mu g.L^{-1}$	[Co] dissolved fraction > 0.1 $\mu g.L^{-1}$	Matching
Threshold DGT n°1	[Co]$_{DGT}$ < 0.16 $\mu g.L^{-1}$	56/74	4/74	81 %
	[Co]$_{DGT}$ > 0.16 $\mu g.L^{-1}$	10/74	4/74	
Threshold DGT n°2	[Co]DGT < 0.04 µg.L-1	8/74	0/74	22 %
	[Co]DGT > 0.04 µg.L-1	58	8/74	

From Tables 6 and 7, it can be seen that interpretation based on dissolved concentration results or DGT results are compliant at 81 % based using Threshold DGT n°1. Among 8 results above the Thresholds marine water, 4 are above the Thresholds DGT. But considering the sites (DEBA, FAL, Saint-Nazaire, Aveiro), only one site (DEBA) is not above the DGT Threshold and the results are very near to both Thresholds DGT n°1 (DGT result: 155 compared to Threshold DGT N°1: 160 ng.L-1), and Threshold marine water (Dissolved fraction result: 108 compared to Threshold marine water: 100 ng.L-1). **So this Threshold $_{DGT}$ n°1 (0.16 $\mu g.L^{-1}$) adapted from Threshold $_{marine\ water}$ (0.1 $\mu g.L^{-1}$) could be considered and proposed for test application in real condition.**

The compliance with Threshold DGT n°2 is only 22%, thus the threshold DGT n° 2 is not appropriate.

Table 7: Co: simulation of data interpretation per site and season using DGT or dissolved fraction results

Site	Season	type	Cobalt DGT results ng.L⁻¹	Interpretation Threshold DGT n°1 0,16 µg.L⁻¹ (160 ng.L⁻¹)	I0nterpretation Threshold DGT n°2 0,04 µg.L-1 (40 ng.L-1)	Cobalt ICPMS results	Interpretation Threshold marine water
DEBA	DS	estuary	155		> 40	108	> 100
LEZO	DS	estuary	29			47	
PRACTICOS	DS	estuary	50		> 40	17	
DEBA	WS	estuary	156		> 40	24	
LEZO	WS	estuary	168	> 160	> 40	68	
MUSEO_D3	WS	estuary	88		> 40	37	
MUSEO_D5	WS	estuary	80		> 40	36	
PRACTICOS	WS	estuary	107		> 40	38	
BELFAST	DS	estuary	42		> 40	54	
FAL_D2	DS	estuary	147		> 40	156	> 100
FAL_D4	DS	estuary	119		> 40	117	> 100
X38A	DS	coastal	33			24	
BELFAST	WS	estuary	171	> 160	> 40	36	
FAL_D2	WS	estuary	238	> 160	> 40	164	> 100
FAL_D4	WS	estuary	114		> 40	161	> 100
LIVERPOOL	WS	coastal	110		> 40	12	
X38A	WS	coastal	110		> 40	5	
ABW_D2	DS	estuary	174	> 160	> 40	44	
ABW_D4	DS	estuary	160		> 40	43	
M69	DS	estuary	99		> 40	20	
M70	DS	estuary	244	> 160	> 40	8	
DUBLINBAYBUOY2	DS	coastal	120		> 40	2	
DUBLINBAYBUOY4	DS	coastal	137		> 40	4	
ABW	WS	estuary	55		> 40	34	
M69	WS	estuary	55		> 40	14	
M70	WS	estuary	70		> 40	8	
DUBLINBAYBUOY4	WS	coastal	53		> 40	14	
FONTENELLE	DS	estuary	61		> 40	9	
TERENEZ	DS	estuary	205	> 160	> 40	81	
BESSIN	DS	coastal	222	> 160	> 40	90	
LAZARET	DS	coastal	72		> 40	13	
SAINTNAZAIRE_D4	DS	coastal	237	> 160	> 40	100	> 100
SAUMONARD	DS	coastal	60		> 40	17	
FONTENELLE	WS	estuary	87		> 40	11	
ANTIFER_BAF	WS	coastal	42		> 40	21	
BESSIN	WS	coastal	119		> 40	41	
LECROISIC	WS	coastal	44		> 40	19	
SAINTNAZAIRE_D2	WS	coastal	214	> 160	> 40	53	
SAINTNAZAIRE_D4	WS	coastal	284	> 160	> 40	54	
SAUMONARD	WS	coastal	243	> 160	> 40	7	
SILLONANGLAIS_BA	WS	coastal	119		> 40	50	
AVEIRO_D3	WS	estuary	293	> 160	> 40	139	> 100
AVEIRO_D5	WS	estuary	216	> 160	> 40	176	> 100
PORTO	WS	coastal	53		> 40	33	
SESIMBRA_D2	WS	coastal	44		> 40	7	
SESIMBRA_D4	WS	coastal	31			9	
TAGUS_D2	WS	coastal	53		> 40	27	
TAGUS_D4	WS	coastal	40			18	
GANDO	DS	coastal	95		> 40	3	
JINAMAR	DS	coastal	208	> 160	> 40	4	
LUZ_D2	DS	coastal	79		> 40	13	
LUZ_D4	DS	coastal	138		> 40	13	
LUZ_2_D4	DS	coastal	126		> 40	25	
TALIARTE	DS	coastal	134		> 40	5	
TALIARTE_2_D2	DS	coastal	148		> 40	7	
TALIARTE_2_D4	DS	coastal	89		> 40	8	
GANDO	WS	coastal	108		> 40	10	
JINAMAR	WS	coastal	49		> 40	8	
LUZ_D3	WS	coastal	128		> 40	22	
LUZ_D7	WS	coastal	46		> 40	23	
TALIARTE	WS	coastal	44		> 40	13	
BRAEHEAD	DS	coastal	151		> 40	63	
MONTROSE	DS	coastal	40			12	
NEWHAVEN	DS	coastal	58		> 40	2	
MOLODOGANA	DS	coastal	55		> 40	31	
MOLOINCHUSA	DS	coastal	138		> 40	24	
MOLORINASCITA	DS	coastal	111		> 40	41	
SANTELMO_D2	DS	coastal	66		> 40	24	
SANTELMO_D5	DS	coastal	52		> 40	21	
MOLODOGANA	WS	coastal	35			24	
MOLOINCHUSA	WS	coastal	52		> 40	18	
MOLORINASCITA	WS	coastal	30			21	
SANTELMO_D3	WS	coastal	155		> 40	20	
SANTELMO_D5	WS	coastal	38			21	

3.3. Copper

The graphical representation of the relationship between concentration of copper measured in dissolved fraction and the labile concentration measured in DGT is presented in Figure 4. The regression line is surrounded by its prediction interval (PI95%), the statistical process (Fig 4 a)) and the determination of Threshold $_{DGT}$ (Fig 4 b)) are presented. For copper, the model validation assumptions were partially satisfied but the relationship between DGT and dissolved concentrations measured by ICP-MS was significant. The model validation criteria were met (p-Values, homoscedasticity, …) with the exception of residue normality, which does not necessarily present a rebuttal aspect to the invalidation of the model, in addition the normality tests p-value (especially Shapiro) is not very far by the threshold of 0.05 (cf Data processing). The relationship between concentration measured in DGT and in dissolved fraction was significant (Fig.4)), and can be described by the equation 4:

$$[Cu]_{DGT} = 0.78\ [Cu]_{Dissolved\ fraction} + 46 \qquad (Eq.\ 4)$$

Concentrations are expressed in $ng.L^{-1}$

The variability in the concentration measured in DGTs is explained at 70 % by the dissolved concentrations measured by ICP-MS. From this model, a concentration of metal in DGT can be predicted from its concentration measured in dissolved fraction. Based on the MONITOOL data, the upper limit of the validity area of this model is around 4,800 $ng.L^{-1}$, the range of validity correspond to the area in light green (Figure 4 (b)). Beyond this limit, this model is not verified.
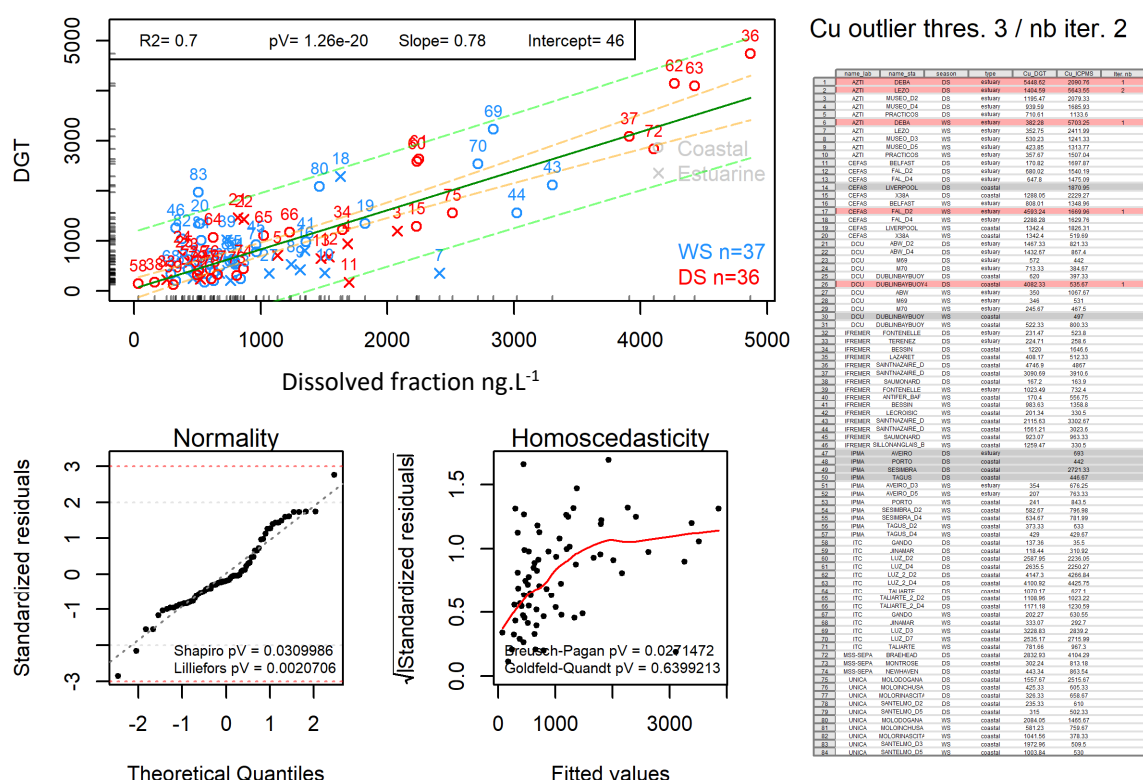
Figure 4 a): Copper: Relationship between dissolved concentration and DGT labile concentration - Statistical process. The dark green line corresponds to the linear model, which is surrounded by its confidence interval CI 95 (orange dashed lines), and its predictione interval (PI 95) (green dashed lines).
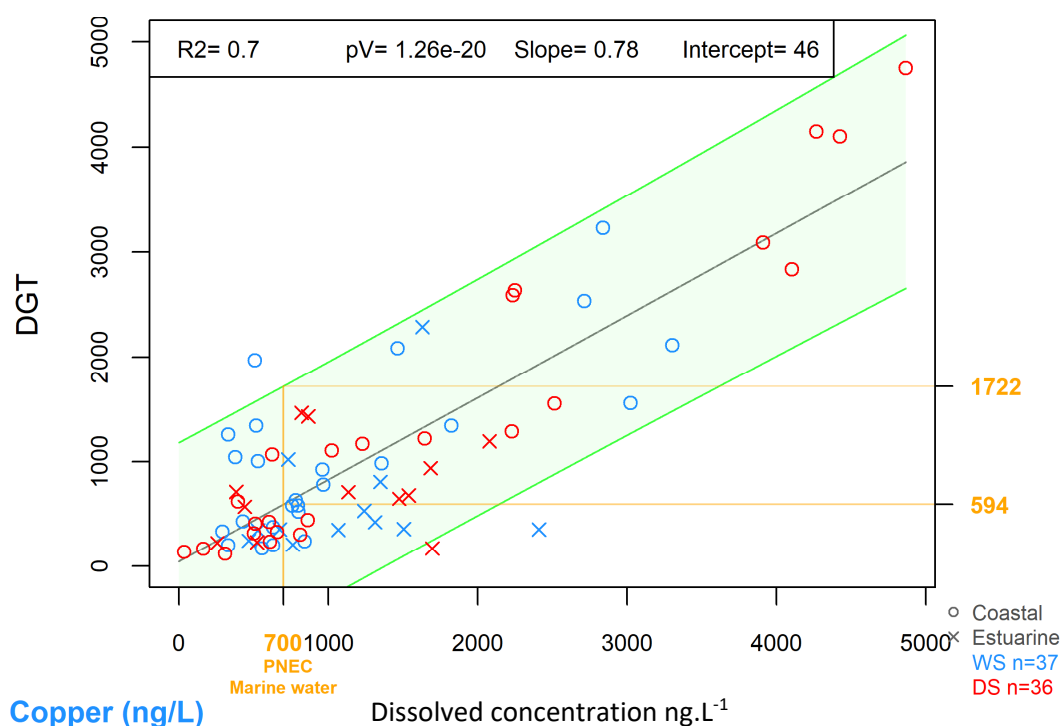
Figure 4 b): Copper: Relationship between concentration in dissolved fraction and in DGT labile concentration - determination of a PNEC $_{DGT}$ – the regression line (black line) is surrounded by its prediction interval (PI 95) in green line in the range of validity (area in light green).

A PNEC $_{marine\ water}$ is defined for copper (0.7 µg.L^{-1}), in the range of validity of the model. The predicted concentration in DGT for this threshold is indicated in Table 7. In order to be more protective, the low value of the prediction interval could have been selected to propose a Threshold $_{DGT}$, but it is not possible as it can not be determined (below the LQ).

Copper is listed as specific pollutant in two of the member states involved in MONITOOL project, thus national thresholds are defined in those countries: 3,8 and 25 µg.L^{-1}. The predicted concentration in DGT for these national thresholds are precised in table 7.

Table 7: Determination of Threshold $_{DGT}$ for copper

Threshold $_{Marine\ water}$ (µg.L^{-1})	Threshold $_{DGT}$ n°1 (µg.L^{-1}) Linear Model Regression	Threshold $_{DGT}$ n°2 (µg.L^{-1}) Linear Model Regression minus low prediction interval (PI 95%)
0.7	0.6	ND (< LQ)
National threshold defined		
3.8	3.16	2.85
25	20.3	19

An overview of the matching is done in Table 8 between assessment based on the dissolved concentration or DGT results. Table 9 presents the detailed results for each site and sampling campaign compared to the

thresholds: DGT results to Thresholds $_{DGT}$ n° 1 and 2 and Dissolved fraction results to the threshold defined in marine water (0.7 µg.L^{-1}).

Table 8: Copper - Matching between assessment based on thresholds defined in marine water and in DGT

		[Cu] dissolved fraction < 0.7 µg.L^{-1}	[Cu] dissolved fraction > 0.7 µg.L^{-1}	Matching
Threshold DGT n°1	[Cu]$_{DGT}$ < 0,6 µg.L^{-1}	20/73	13/73	71 %
	[Cu]$_{DGT}$ > 0,6 µg.L^{-1}	8/73	32/73	

Thresholds marine water defined at national level				
		[Cu] dissolved fraction < 3.8 µg.L^{-1}	[Cu] dissolved fraction > 3.8 µg.L^{-1}	
Threshold DGT n°1	[Cu]$_{DGT}$ < 3.16 µg.L^{-1}	67/73	2/73	96 %
	[Cu]$_{DGT}$ > 3.16 µg.L^{-1}	1/73	3/73	
		[Cu] dissolved fraction < 25 µg.L^{-1}	[Cu] dissolved fraction > 25 µg.L^{-1}	
Threshold DGT n°1	[Cu]$_{DGT}$ < 20.3 µg.L^{-1}	73/73	0/73	100 %
	[Cu]$_{DGT}$ > 20.3 µg.L^{-1}	0/73	0/73	

		[Cu] dissolved fraction < 3.8 µg.L^{-1}	[Cu] dissolved fraction > 3.8 µg.L^{-}	
Threshold DGT n°2	[Cu]$_{DGT}$ < 2.85 µg.L^{-1}	67/73	1/73	97 %
	[Cu]$_{DGT}$ > 2.85 µg.L^{-1}	1/73	4/73	
		[Cu] dissolved fraction < 25 µg.L^{-1}	[Cu] dissolved fraction > 25 µg.L^{-1}	
Threshold DGT n°2	[Cu]$_{DGT}$ < 19 µg.L^{-1}	73/73	0/73	100 %

From Tables 8 and 9, it can be seen that considering:

- Threshold DGT n°1: interpretation based on dissolved fraction results or DGT results are compliant at 71% and in case of mismatching, DGT threshold is not enough protective compare to Threshold marine water (13 results with results [Cu] dissolved fraction > 0,7 µg.L^{-1} are < DTG thresholds). Threshold DGT n°1 has 29% of missmaching

(Table 10), it can't be proposed directly. Considering that 7 sites are above the Threshold $_{\text{marine water}}$ but below the Threshold $_{\text{DGT}}$ n°1 (most of these sites have only one result aquired in wet season).

- Threshold $_{\text{DGT}}$ n°2 is unapplicable.

- Concerning the 2 Thresholds $_{\text{marine water}}$ defined at national level, the information of matching and mismaching are precised in Table 8. As these thresholds are much higher than the Threshold $_{\text{marine water}}$ "selected", none or only few MONITOOL results are near or above these Thresholds $_{\text{marine water}}$. Thus the matching is near or equal to 100 %.

Table 9: Cu: simulation of data interpretation per site and season using DGT or dissolved fraction results. Threshold $_{marine\ water}$= 0.7 µg.L^{-1}, Thresholds $_{DGT}$ n°1 = 0.6 µg.L^{-1} and n°2 = < LQ.

Site	Season	Copper DGT results ng.L^{-1}	Interpretation Threshold DGT n°1 0,6 µg.L^{-1} (600 ng.L^{-1})	Interpretation Threshold DGT n°2 < LQ	Copper Dissolved fraction results ng.L^{-1}	Interpretation Threshold marine water 0.7 µg.L^{-1} (700 ng.L^{-1})
MUSEO_D2	DS	1195	> 600	> LQ	2079	> 700
MUSEO_D4	DS	940	> 600	> LQ	1686	> 700
PRACTICOS	DS	711	> 600	> LQ	1134	> 700
LEZO	WS	353		> LQ	2412	> 700
MUSEO_D3	WS	530		> LQ	1241	> 700
MUSEO_D5	WS	424		> LQ	1314	> 700
PRACTICOS	WS	358		> LQ	1507	> 700
BELFAST	DS	171		> LQ	1698	> 700
FAL_D2	DS	680	> 600	> LQ	1540	> 700
FAL_D4	DS	648	> 600	> LQ	1475	> 700
X38A	DS	1288	> 600	> LQ	2229	> 700
BELFAST	WS	808	> 600	> LQ	1349	> 700
FAL_D4	WS	2288	> 600	> LQ	1630	> 700
LIVERPOOL	WS	1342	> 600	> LQ	1826	> 700
X38A	WS	1342	> 600	> LQ	520	
ABW_D2	DS	1467	> 600	> LQ	821	> 700
ABW_D4	DS	1433	> 600	> LQ	867	> 700
M69	DS	572		> LQ	442	
M70	DS	713	> 600	> LQ	385	
DUBLINBAYBUOY2	DS	620	> 600	> LQ	397	
ABW	WS	350		> LQ	1068	> 700
M69	WS	346		> LQ	531	
M70	WS	246		> LQ	468	
DUBLINBAYBUOY4	WS	522		> LQ	800	> 700
FONTENELLE	DS	231		> LQ	524	
TERENEZ	DS	225		> LQ	259	
BESSIN	DS	1220	> 600	> LQ	1647	> 700
LAZARET	DS	408		> LQ	512	
SAINTNAZAIRE_D2	DS	4747	> 600	> LQ	4867	> 700
SAINTNAZAIRE_D4	DS	3091	> 600	> LQ	3911	> 700
SAUMONARD	DS	167		> LQ	164	
FONTENELLE	WS	1023	> 600	> LQ	732	> 700
ANTIFER_BAF	WS	170		> LQ	557	
BESSIN	WS	984	> 600	> LQ	1359	> 700
LECROISIC	WS	201		> LQ	331	
SAINTNAZAIRE_D2	WS	2116	> 600	> LQ	3303	> 700
SAINTNAZAIRE_D4	WS	1561	> 600	> LQ	3024	> 700
SAUMONARD	WS	923	> 600	> LQ	963	> 700
SILLONANGLAIS_BAF	WS	1259	> 600	> LQ	331	
AVEIRO_D3	WS	354		> LQ	676	
AVEIRO_D5	WS	207		> LQ	763	> 700
PORTO	WS	241		> LQ	844	> 700
SESIMBRA_D2	WS	583		> LQ	797	> 700
SESIMBRA_D4	WS	635	> 600	> LQ	782	> 700
TAGUS_D2	WS	373		> LQ	633	
TAGUS_D4	WS	429		> LQ	430	
GANDO	DS	137		> LQ	36	
JINAMAR	DS	118		> LQ	311	
LUZ_D2	DS	2588	> 600	> LQ	2236	> 700
LUZ_D4	DS	2636	> 600	> LQ	2250	> 700
LUZ_2_D2	DS	4147	> 600	> LQ	4267	> 700
LUZ_2_D4	DS	4101	> 600	> LQ	4426	> 700
TALIARTE	DS	1070	> 600	> LQ	627	
TALIARTE_2_D2	DS	1109	> 600	> LQ	1023	> 700
TALIARTE_2_D4	DS	1171	> 600	> LQ	1231	> 700
GANDO	WS	202		> LQ	631	
JINAMAR	WS	333		> LQ	293	
LUZ_D3	WS	3229	> 600	> LQ	2839	> 700
LUZ_D7	WS	2535	> 600	> LQ	2716	> 700
TALIARTE	WS	782	> 600	> LQ	967	> 700
BRAEHEAD	DS	2833	> 600	> LQ	4104	> 700
MONTROSE	DS	302		> LQ	813	> 700
NEWHAVEN	DS	443		> LQ	864	> 700
MOLODOGANA	DS	1558	> 600	> LQ	2516	> 700
MOLOINCHUSA	DS	425		> LQ	605	
MOLORINASCITA	DS	326		> LQ	659	
SANTELMO_D2	DS	235		> LQ	610	
SANTELMO_D5	DS	315		> LQ	502	
MOLODOGANA	WS	2084	> 600	> LQ	1466	> 700
MOLOINCHUSA	WS	581		> LQ	760	> 700
MOLORINASCITA	WS	1042	> 600	> LQ	378	
SANTELMO_D3	WS	1973	> 600	> LQ	510	
SANTELMO_D5	WS	1004	> 600	> LQ	530	

3.4. Manganese

The graphical representation of the relationship between concentration of manganese measured in dissolved fraction and the labile concentration measured in DGT is presented in Figure 5. The regression line is surrounded by its prediction interval (PI95%), the statistical process (Fig 5a) and the determination of Threshold DGT (Fig 5b) are presented. For manganese, the model validation assumptions were partially satisfied but the relationship between concentration measured in DGT and in dissolved concentrations was significant. The model validation criteria were met (p-Values, homoscedasticity, …) with the exception of residue normality, which does not necessarily present a rebuttal aspect to the invalidation of the model (cf Data processing). The relationship between DGT and dissolved concentration was significant (Fig.5), and can be described by the equation 5:

$$[Mn]_{DGT} = 1.53\,[Mn]_{Dissolved\ fraction} + 1883 \qquad (Eq.5)$$

Concentrations are expressed in $ng.L^{-1}$

The variability in the concentration measured in DGTs is explained at 91% by the dissolved concentrations measured by ICP-MS. From this model, a concentration of metal in DGT can be predicted from its concentration measured in dissolved fraction. Based on the MONITOOL data, the upper limit of the validity area of this model is around 25,000 $ng.L^{-1}$, the range of validity correspond to the area in light green (Figure 6 (b)). Beyond this limit, this model is not verified.

Figure 5 a): Manganese: Relationship between dissolved concentration and DGT - Statistical process. The dark green line corresponds to the linear model, which is surrounded by its confidence interval CI 95 (orange dashed lines), and its prediction interval (PI 95) (green dashed lines).
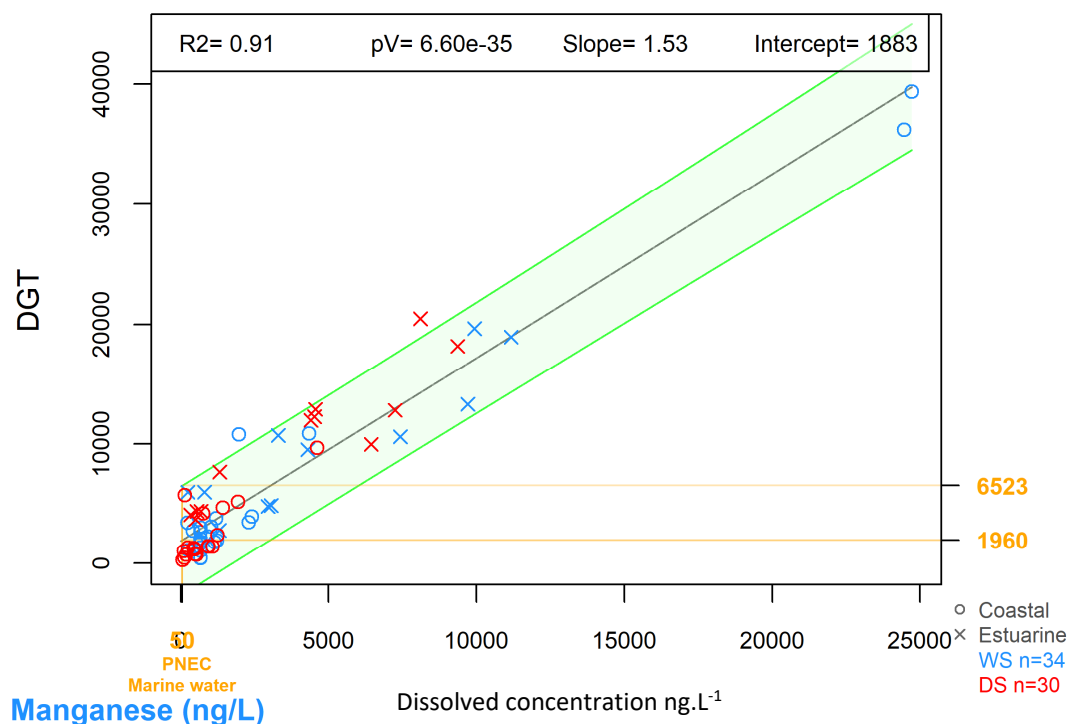
Figure 5 b): Manganese: Relationship between dissolved concentration and DGT - determination of one Threshold$_{DGT}$ – the regression line (black line) is surrounded by its prediction interval (PI 95) in green line in the range of validity (area in light green).

One threshold is defined for manganese, it is a PNEC $_{add\ marine\ water}$, the predicted concentration in DGT is precised in table 10.

Manganese is listed as specific pollutant in one of the member states involved in MONITOOL project, thus national threshold is defined in this country: 123 µg.L^{-1}. The predicted concentration in DGT for this national threshold is precised in Table 10.

Table 10*: Determination of Threshold $_{DGT}$ for manganese

Threshold $_{Marine\ water}$ (µg.L^{-1})	Threshold add $_{DGT}$ n°1 (µg.L^{-1}) Linear Model Regression	Threshold add $_{DGT}$ n°2 (µg.L^{-1}) Linear Model Regression minus low prediction interval (PI 95%)
0.5	2	<LQ
National threshold defined		
123	190	182

23

An overview of the matching is done Table 11 between assessment based on the dissolved concentration or DGT results. 73% of the assessment are matching (Table 11) but as it is a threshold "add" the use of this threshold requires to be added to the background concentration to be interpreted, thus it should be considered in relation to the site (at a local stage) and knowing the background level. Further data interpretation (related to the background level) site specific are thus necessary to assess if this threshold adaptation is appropriate and should be considered. **It could be proposed to test the Threshold $_{add\ DGT}$ for manganese, in order to check his applicability in real condition.**

Concerning the Threshold $_{marine\ water}$ defined at national level, the information of matching and mismaching is precised in Table 11. As this threshold is much higher than the Threshold marine water "selected", none MONITOOL results are near or above these Thresholds $_{marine\ water}$. Thus the matching is equal to 100%.

Table 11: Manganese - Matching between assessment based on thresholds defined in marine water and in DGT

		[Mn] dissolved fraction < 0,5 µg.L^{-1}	[Mn] dissolved fraction > 0,5 µg.L^{-1}	Matching
Threshold DGT n°1	**[Mn]$_{DGT}$ < 2 µg.L^{-1}**	9/64	12/64	73 %
	[Mn]$_{DGT}$ > 2 µg.L^{-1}	5/64	38/64	

Threshold marine water defined at national level				
		[Mn] dissolved fraction < 123 µg.L^{-1}	[Mn] dissolved fraction > 123 µg.L^{-1}	
Threshold DGT n°1	**[Mn]$_{DGT}$ < 190 µg.L^{-1}**	64/64	0	100 %
Threshold DGT n°2	**[Mn]$_{DGT}$ < 182 µg.L^{-1}**	64/64	0	100 %

3.5. Zinc

For zinc, the acceptance criteria of the model are not fully met (normality is not really good) but above all the concentrations measured in the DGTs are explained by only 6% of the model (Figure 6). These elements are not satisfactory to adapt a DGT threshold from the threshold marine water for zinc (based on MONITOOL data).

Figure 6: Zinc: Relationship between dissolved concentration and DGT - Statistical process. The dark green line corresponds to the linear model, which is surrounded by its confidence interval CI 95 (orange dashed lines), and its predictione interval (PI 95) (green dashed lines).

Conclusion

The adaptation of EQS $_{DGT}$ for cadmium, nickel and lead was done in the action 1 (6). As the analysis of DGT (Chelex® 100 resin) can provide results for several other metals: cobalt, copper, manganese, zinc and iron, the adaptation of existing thresholds for these metals was carried out. A survey between MONITOOL partners and associated partners pointed out that among these five metals, four are identified as specific pollutants in one or two member states: copper, zinc, iron, and manganese.

A search of existing thresholds was done for these metals (cobalt, copper, manganese, zinc and iron), on different database. After a first assessment of the robustness of existing thresholds, based on one expert judgment, a threshold for each metal was "selected" for MONITOOL, so that an adaptation of this threshold to DGT threshold can be done.

In addition, when a metal is identified as specific pollutant by a member state, this has in charge to determine a national threshold. Thus, national thresholds are also defined for copper, zinc, iron and manganese by one or two member states. These thresholds were also considered.

The adaptation of a metal threshold defined in marine water to threshold DGT implies to establish for each considered metal a valid relationship between the dissolved concentration measured in spot water samples and the DGT labile concentration measured in DGTs.

Based on MONITOOL result, a statistical process was run for cobalt, copper, manganese and zinc.

Among these metals, only copper and manganese are identified specific pollutant for one or two member states involved in MONITOOL.

For cobalt, Threshold DGT n°1 (0.16 µg.L^{-1}) adapted from Threshold marine water (0.1 µg.L^{-1}) could be considered and proposed for test application in real conditions.

For copper, Threshold DGT n°1 (0.6 µg.L^{-1}) adapted from Threshold marine water (0.7 µg.L^{-1}) appears not enough protective compared to the threshold marine. Nevertheless, national Thresholds defined in marine water (3.8; 25 µg.L^{-1}) are much higher and thus, adapted Threshold DGT (3.16; 20,3 µg.L^{-1}) can be determined. At these concentration levels, no mismatching between assesment of results based on dissolved fraction (compare to Threshold marine water) or DGT (compare to DGT Threshold) can be pointed out based on MONITOOL data.

For zinc, no DGT threshold can be adapted based on MONITOOL data, due to the poor relationship existing between concentration measured in dissolved fraction (spot water samples) and in DGT.

For manganese, the selected threshold in marine water is defined as a "Threshold add", this means that to carry out an assessment, it should be considered a site specific approach, the assessment shoud be done considering the background concentration level:

$$\text{PNEC}_{\text{marine water}} = \text{PNEC}_{\text{add marine water}} + \text{background concentration}$$

Thus, in order to assess the relevancy of the Threshold add DGT, further assessment with site specific approach should be considered, this would need to considere the background concentrations. DGT should be considered in this way, as it could be a very useful tool to access the background concentration (7).

Bibliography

1. E.C. 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. Official Journal of the European Union L327/1, 22.12.2000.

2. E.C. Final draft revised Guidance Document No.27. Technical Guidance for deriving Environmental Quality Standards. June 2018.

3. Bolam T, Bersuder P. Monitool- Sampling protocol. Cefas. 20p. 2018.

4. Knief U, Forstmeier W. Violating the normality assumption may be the lesser of two evils. bioRxiv [Internet]. 2020 May 5 [cited 2020 Sep 28];498931. Available from: https://www.biorxiv.org/content/10.1101/498931v2

5. Wasserstein RL, Schirm AL, Lazar NA. Moving to a World Beyond " p < 0.05". The American Statistician [Internet]. 2019 Mar 29 [cited 2020 Sep 28];73(sup1):1–19. Available from: https://www.tandfonline.com/doi/full/10.1080/00031305.2019.1583913

6. Amouroux I, Gonzalez J-L, Guesdon S, Menet F, Dallet M. DGT: EQS adaptation for Nickel, Cadmium and Lead. MONITOOL project. 2021.

7. Marras B, Montero N, Marrucci A, Bettoschi A, Atzori M. Operational DGT threshold values for metals in seawater from protected coastal areas in Sardinia (Western Mediterranean). Marine Pollution Bulletin. volume 150. 2020 Jan;