Marine Life Information Network
Marine Biological Association of the United Kingdom

MARLIN – MARINE LIFE INFORMATION NETWORK

SENSITIVITY ASSESSMENT OF CONTAMINANT PRESSURES

RAPID EVIDENCE ASSESSMENT (REA) - PROTOCOL

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1 Rapid Evidence Assessment (REA) - Protocol

The 'Contaminants' Rapid Evidence Assessment (REA) protocol was designed to collate and synthesis the evidence required to inform the assessment of the resistance to, and hence sensitivity of, marine habitats and species to the MarESA 'Contaminant' pressures. These are 'Hydrocarbons and PAHs'; 'Transition elements and organo-metals'; 'Synthetic compounds (inc. pesticides, antifoulants, pharmaceuticals)' and 'Introduction of other substances (solid, liquid, gas)'.

1.1 Background

MarESA resistance assessment is based on the evidence collated in the literature review on the effects of each pressure (or activity that results in a given pressure) on the key elements of the feature (physical habitat and species that contribute to sensitivity). Resistance assessment considers the following for each pressure in turn:

- reported evidence on the direct effect of a given pressure on the key elements of the feature, compared to the benchmark level of pressure;
- the resultant levels of damage on the key elements, e.g. extent of damage to habitat, loss of population size or abundance, changes in diversity, loss or reduction in abundance of one of more species groups;
- reported evidence on the direct effect of a given pressure on similar habitats, species, or functional groups, and/or
- 'proxies' are used to inform the assessment of the likely effect of a pressure on the key elements of the feature, in the absence of direct evidence.

Wherever possible, direct evidence of the effect of a given pressure on the 'key elements of the feature' (habitat and/or the species) is used as the basis of the assessment of resistance. Where the evidence quantifies the magnitude, extent or frequency of the pressure then the evidence can be compared directly with the benchmark. Similarly, if the pressure is qualified in the evidence then it can be compared with the relevant benchmark. The quality of the evidence and its applicability to each pressure assessment is ranked using the 'confidence assessment' scale (Tyler-Walters *et al.*, 2018).

In some cases, where evidence is lacking, it is possible to use 'proxies' against which a resistance assessment can be made. For example, congeners or members of the same taxonomic Class or even Phylum may be suitable 'proxies' for the physiological or toxicological effects of one or more chemical groups. Similarly, chemicals that have the same mode of action or act on the same metabolic pathway may be proxies for other chemicals that are not studied in detail.

The resultant 'resistance' assessments are combined with a species or habitat 'resilience' assessment, reviewed separately, to determine an overall sensitivity assessment (Tyler-Walters *et al.*, 2018).

Phase 1 of the 'contaminants' project concluded that quantified benchmarks were impractical so that a 'weight of evidence' approach was the most practical way to assess resistance to the effects of contaminants on marine habitats and their species. It also concluded that the REA approach was a useful approach to improve and standardise the literature review process.

1.2 Current REA protocol

The evidence review process was based on the Defra/NERC Quick Scoping Reviews and Rapid Evidence Assessments guidance[1] (Collins *et al.*, 2015) together with examples of relevant REAs and Systematic

[1] https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/560521/Production_of_quick_scoping_reviews_and_rapid_evidence_assessments.pdf

reviews (Johnston & Roberts, 2009; Johnston *et al.,* 2015; Randall *et al.*, 2015; Collier *et al.*, 2016; Mayer-Pinto *et al.,* 2020).

The REA process involves the following steps (summarized from Collins *et al.*, 2015).

- Develop protocol (including the details of the evidence review question(s) and methodology)
- Search for evidence (using the search strategy and methodology in the protocol)
- Screen the search results using relevancy (inclusions and exclusion) criteria outlined in the protocol
- Extract evidence relevant to the evidence review question(s) – and create a 'map of the evidence'
- Critically appraise the evidence and its relevance to the 'review question'.
- Synthesize the results.
- Communicate the results

1.3 Defining of the 'review question'

The 'Contaminants' (REA) review aimed to provide the information required to assess the likely effect of any given 'contaminant' pressure on a range of marine habitats and their associated species.

MarESA resistance assessment is based on effects that result in:

- the loss of or reduction in population size, extent, or abundance of one or more species groups within the habitat,
- the loss of diversity, and/or
- damage to the extent or function of the habitat (see above).

These are likely to result from:

- the direct mortality of adults and their loss from the habitat/species population,
- the direct mortality of larvae, juveniles or other propagules so that recruitment is reduced/prevented, or
- direct or indirect effects on reproduction and recruitment resulting in population decline.

Resistance assessment, in MarESA, is predicated on evidence of 'mortality', 'population decline, and/or habitat modification. In most cases, it is assumed that the 'contaminant' pressures will affect habitats via their effects on individual species. The exceptions are the physical effects of oils and the ecosystem-wide effects of nutrient enrichment.

Therefore, the evidence requirements can be expressed as the following **'review question'**:

'Does exposure of taxon 'a' to contaminant 'x' result in:

1. **the direct mortality of adults and their loss from the habitat or population,**
2. **the direct mortality of larvae, juveniles or other propagules so that recruitment is reduced/prevented,**
3. **or direct or indirect effects on reproduction and recruitment resulting in population decline of one or more species in the habitat of interest.**

Therefore, the 'Contaminants' REA evidence reviews concentrate on the evidence required to answer the above question. The term 'taxon' is used to denote the relevant taxonomic level of unit, e.g. Species, Genus, Family, Order, Class, or Phylum.

1.4 Scope (inclusions and exclusions)

The proposed scope is outlined below and summarized in the following PICO table (Table 1.1).

1. **Marine benthic habitats** - include all (ca 400+ biotopes) identified by the UK Marine Habitat Classification (UKMHC; JNCC, 2015) from the supralittoral to the sublittoral including habitats in the shallow shelf seas (<200 m deep) and deep-sea (>200 m deep) (Connor *et al.*, 2004; Parry *et al.*, 2015; JNCC, 2015).
2. The focus is on marine, estuarine and transitional water habitats as listed in the UKMHC (JNCC, 2015)
3. **Marine benthic species** refers to the agreed list of species 'indicative of sensitivity'[2] (**Error! Reference source not found.**), together with their congeners, co-familial and/or members of the same taxonomic group including Class and Phylum where required.
4. **Geographic range** –United Kingdom habitats and species, although relevant information from the coasts of the North East Atlantic and other temperate regions will be included where required. Where evidence is lacking, information from similar species and habitats in the temperate southern seas, or tropics will be included.
5. **Biogeographic range** – marine temperate species and habitats in the northern hemisphere. Where evidence is lacking, information from similar species and habitats in the temperate southern seas, or tropics will be included.

Information on the effects of contaminants on many marine species may be poorly studied. Therefore, the species range may be expanded to include similar congeners, members of the same family or Phylum. At present the emphasis in on marine benthic species and their propagules/larvae/juveniles. Therefore, demersal and pelagic mobile species are excluded, in particular, fish, marine reptiles and marine mammals. Phyto- and Zooplankton are also excluded, except where they include the larval or juvenile stags of the benthic species of interest.

The biogeographic and geographic range may be expanded to include evidence from the southern hemisphere and/or tropics. The emphasis is on UK marine and brackish water species and habitats. Freshwater species are excluded except where they can be used a 'proxies' for species of the same taxonomic group or that share the same AOP/MIE for one or more chemicals of interest.

6. **'Contaminant'** refers to those groups of chemicals and individual chemicals listed in the agreed table of contaminants (Contaminant Chemicals Groups' March 2022 spreadsheet). At present, the agreed list excludes macro-plastics, micro-plastics and other marine debris. Chemicals that evaporate if spilt (evaporators) are also excluded.
7. **'Nutrients'** and 'organic enrichment' are excluded because these pressures have already been subject to MarESA sensitivity assessment.
8. **'Exposure'** - the following potential routes of 'exposure' to contaminants are included**:**

- physical contact – e.g. smothering/clogging by oils;
- physical ingestion – e.g. of oils or particulates;
- ingestion and/or absorption from water i.e. the water column or interstitial water;
- ingestion /absorption from food including contaminants adsorbed onto organic or inorganic particulates, or
- absorption from the substratum e.g. sediment.

[2] As defined in the MarESA Guidance Manual 2018 (Tyler-Walters *et al.*, 2018).

At present, inhalation by birds, reptiles, or mammals is excluded, as these mobile species are not included in the study.

Table 1.1. PICO elements and summary of relevant inclusion and exclusion criteria.

		Inclusion criteria	Exclusion criteria
Population	Marine benthic habitats and their component species	All species on agreed list, plus congeners, co-familial Members of same taxonomic groups e.g. Order, Class, Phyla	Mammals, reptiles, birds, fish; phytoplankton (unless a macroalgal propagule); viruses; zooplankton (unless a relevant larval stage).
Intervention (Exposure)	Physical smothering/ingestion/clogging Ingestion/absorption via water Ingestion/absorption via sediment/substratum Ingestion/absorption via food	Agreed list of chemicals Nanoparticulate / Engineered Nanomaterials	Air borne gases Evaporators that disperse at water surface Plastics/Microplastics Non-toxic spills – e.g. coal, wheat, grains etc.
Comparator	Examination of the effect of a contaminant, compared to a control on a species or habitat of interest Examination of the effect of a contaminant before or after a spill or incidental release into a habitat or on a species population of interest	Quantitative experimental controlled laboratory studies inc. randomized control and non-randomized control studies Quantitative experimental, controlled, in situ (field) studies/survey inc. randomized control and non-randomized control studies Quantitative observational studies/survey of before and after spills/incidents, case-controls Quantitative observational studies of long-term effects Quantitative or qualitative reviews – literature reviews, systematic reviews.	Anecdotal observations
Outcome	**Species** Toxicity (mortality of adult/larval/propagule)	Direction of effect (i.e. increase or decrease)	Accumulation studies e.g. bioaccumulation, bioindicator studies (except if they explain/result in

		Inclusion criteria	Exclusion criteria
	Larval/juvenile abnormalities Physical (smothering, suffocation, clogging) Toxicity (reproductive impairment; endocrine disruption) Toxicity (effect on growth, repair) Behavioural response (resulting in population decline (e.g. due to mating failure), feeding behaviour resulting in mortality or reduced fecundity/recruitment or loss from site of interest) **Habitat** Physical/chemical habitat modification resulting in recruitment failure Physical modification of the habitat (smothering) Change in species diversity, population extent, species abundance and community composition (i.e. biotope) Changes in trophic interactions (e.g. abundance of grazers), productivity,	Qualification or quantification of effect Lethal effect concentrations (e.g. LC_{50}, PEC) No-effect concentrations (e.g. NOEC, PNEC) Sub-lethal effects	mortality and population decline) Biochemical (except if they explain/result in mortality and population decline) Cellular/molecular studies (transomics and genomics) (except if they explain/result in mortality and population decline) Physiology or behaviour (except if they explain/result in mortality and popn decline) Studies of population genetics, ecology, autoecology, taxonomy, socio-economics, or non-contaminant –based 'pressures'

9. **Maritime activities –** the review includes operational and incidental spills, operational releases, and discharges from maritime activities (offshore and inshore), as well as activities that discharge into water courses that ultimately reach marine waters (Table 1.2).

The review will prioritize releases in the marine environment but will also need to include freshwater (riverine) inputs. Exposure from aerosol deposition is excluded except where the aerosol is known to dissolve in water and becomes available to benthic species.

1.5 Search for evidence

Peer-reviewed and grey literature is searched using:

- Web of Science (WoS; Core Collection: Citation Indexes) 1970-present, and

- US EPA's ECOTOX[3] database.

In addition:

- Review articles, including systematic reviews, are used to speed up the literature review where possible.
- The SCOPUS bibliographic database is not used, unless no other database reveals results.
- Google Scholar is used to fill gaps, e.g. in grey literature, where other databases reveal few 'hits', that is, in otherwise poorly studied group and species.

Additional evidence will be obtained from the references lists of the literature discovered, together with relevant review articles and reports.

Table 1.2. Maritime/Coastal activity and process-based search terms (preliminary).

Primary term	**Secondary terms**
Shipping (commercial & recreational)	Vessels / Tankers / Freight
Spills (Accidental/Incidental)	Shipping (commercial & recreational) Inshore/offshore installations Harbours/Ports/Berths/Moorings/Bunkering Chemical spills Oil spills Spills of containers/barrels (oil and chemical)
Operational discharges	Shipping (commercial & recreational) Bilge water Ballast tanks Oil & gas installations Renewables/wind farms (inc. cable installation, support vessels) Mariculture Harbours/Ports/Berths/Moorings/Bunkering
Antifouling paints	Shipping (commercial & recreational) Mariculture/Aquaculture/Fin and shellfish) Offshore renewable Harbour/Port infrastructure (e.g. buoys pontoons etc.)
Mariculture/Aquaculture (fin-fish, shellfish)	Chemotherapeuticals/Medicines Antibiotics Parasiticides/Biocides Anaesthetics Disinfectants Food supplements Antifoulants
Oil & gas exploration/ production	Drilling wastes/muds/cuttings Produced waters Decommissioning Cables/Pipelines
Inshore/Offshore renewable (inc. cables)	Antifoulants Construction/decommissioning Sediment remobilization

[3] US EPA Ecotoxicology database- https://cfpub.epa.gov/ecotox/

Primary term	Secondary terms
Dredging and dumped spoil	Aggregate dredging Channelization Harbours/Ports
Inshore discharges/outfalls	Power stations Industrial effluents Sewerage effluents (inc. human pharmaceuticals)
Runoff	Agricultural runoff (e.g. hormones/pesticides/nutrients) Urban runoff Mine effluents/waste runoff
Munitions dumps	Chemical warfare agents Explosives/propellants
Ship wrecks	

Additional information on toxicology will be obtained from the HNS database and/or the AOP wiki. The Marine Biological Association's (MBA) library catalogue will be used to fill gaps, especially in grey literature and literature prior to 1980, if needed.

A date range of '1970 to present' is used. However, the range may be extended to 1960s for information on oil spills and experimental oil spills. The MarLIN Steering Group or relevant MBA experts will be approached for relevant un-published evidence, where required.

Key words and search strings

Key words based on the pressure name, contaminant groups, contaminant type (Section 2.6; 'Contaminant Chemicals Groups' March 2022 spreadsheet), and marine activities list (see Additional evidence will be obtained from the references lists of the literature discovered, together with relevant review articles and reports.

Table 1.2) were used to develop a suite of standard 'search strings' against each taxon (Appendix 2).

Each of the search strings developed for taxa or habitat type (Appendix 2) were applied in turn across each of the bibliographic databases examined (WoS, SCOPUS, Google Scholar). The search for evidence is designed to be as inclusive as possible so that no potential source of evidence is overlooked. However, we used common sense to determine the best search strings to use.

For example, if the least restrictive search string gave an unmanageable number of 'hits' (e.g. several hundred) then a more restrictive (focused) string was used. Conversely, if the number of 'hits' was very low (e.g. <10) or zero then a less restrictive search string was used (see spreadsheet). Some species resulted in zero hits for most searches. In these cases, (e.g. the sea pens*, Leptometra, Ocnus* etc.) then the broadest search (ALL=Taxon) was used to capture everything listed for that species in the bibliographic database.

ECOTOX has its own search tools that enable the user to specify individual species or broad taxonomic groups (e.g. Molluscs), chemicals, effects, and 'end points'. But, the ECOTOX is a specialist database that records available toxicological information from a wide range of species and habitats so that the majority of 'hits' obtained in ECOTOX are directly relevant to the 'review question'. The following search parameters were used in the ECOTOX database:

- Species name– taxon name, and/or
- Species group (e.g. Molluscs, Crustaceans, etc.) if required to broaden the search
- Chemicals – set to 'All'

- Endpoints – set to 'All'
- Publications - – set to 'All'
- Test conditions – set to 'All'
- Effect groups –, 'Biochemical', 'Cellular', 'Behavioural', 'Ecosystem', 'Growth', 'Multiple', 'Mortality', 'Physiology', 'Population', and 'Reproduction' groups are included but the 'Accumulation' group is excluded.

Search results

The number of the search results (hits) is recorded together with the date of the literature review. The resultant citations were downloaded from the relevant bibliographic database into Endnote (X20.1). The results of the ECOTOX searches were downloaded in Excel format, dated, and copied into the relevant 'Evidence summary' spreadsheet.

1.6 Screening

The resultant list of articles was then subject to a two stage screening process (Collins *et al.,* 2015) against the inclusion and exclusion criteria.

1. Stage 1 involved a look at the title the articles and a quick look at the abstract.
2. Stage 2 involved an examination of the abstract, introduction, and possibly conclusions of the articles based on a speed-reading of the article.

Stage 1 is intended to remove articles captured by the search strings that are obviously not relevant to the study. Stage 2 screening is intended to exclude those articles that are definitely not relevant to the 'review question'.

Collins *et al.* (2015) recommended that the results of Stage 2 were recorded together with reasons why each article was included or excluded based on the inclusion/exclusion criteria. However, this record was omitted due to the time constraints.

The following types of articles were included during screening:

- Papers that examine effects (sub-lethal, lethal, population) of one/more contaminant on the species or habitat of interest;
- Papers that examine effects (sub-lethal, lethal, population) of one/more contaminant on another similar species where no information on specific contaminants on the species of interest was found;
- Papers that might be relevant or link to relevant evidence but are unclear from title/abstract, or only title available;
- Review articles that pointed to other relevant evidence; and
- Evidence of sub-lethal effects on reproduction/scope of growth as it has the potential for population decline.

The following exclusions were made during screening:

- Methodological papers e.g. design of assays, biomarkers and their application;
- Metabolic/proteomics/genomics of the effects of chemicals;
- Marine biotoxins – i.e. from algal blooms/HABs;
- Novel chemicals of pharmaceutical potential extracted from species of interest;
- Human pathogens (e.g. *E.coli*, Strep and viruses) accumulated by mussels;

- Articles not relevant the taxon or habitat of interest – unless they were the only mention of chemical of interest in dataset and may function as 'proxies';
- Evidence on effects of shellfish poisoning or shellfish contamination on humans;
- Faecal pollution; and
- General physiology or genetics i.e. not related to the effect of contaminants.

1.7 Evidence extraction, mapping and appraisal

The evidence extracted (or mapped) is limited to fields likely to be relevant to sensitivity assessment or to categorise the 'level of effect' recorded in each article. The extensive systematic map suggested by Randall *et al.* (2015) was felt to be too onerous.

1.7.1 Evidence summary – terms and definitions

The field names and standard terms used within the 'Evidence summaries' were developed during Phase 2 and 3, based on terms used by the US EPA ECOTOX database or MarLIN glossary, or adapted from the literature review, wherever possible or relevant. Not reported (NR) is used wherever the relevant data/evidence is not reported or specified in the evidence. The field names and relevant standard terms follow.

Short citation

Standard short form of citation for article/paper/book/ report etc.

Study type

Outline of the type of study adapted from ECOTOX definitions:

Term	Definition
Field (obs)	Observation in the field e.g. effect of spills, physical disturbance
Field (expt)	Field based study, e.g. in situ mesocosm, field based experimental design exposed and control plots/quadrats/transects
Laboratory	Experimental or observational study conducted under laboratory conditions
Mesocosm	Experimental or laboratory studies conducted within mesocosms either based in the laboratory or the field
Review	Review article (paper/report). Reviews used as sources of evidence and only novel data in reviews included, originals articles examined for detail
Survey	Survey of multiple site presence/absence/abundance etc. of chemical or species

Note –chemical analysis requires access to a laboratory but is not included within the study type.

Chemical names and groups

'Contaminants group', 'contaminant type', 'contaminant name' and 'CAS number' from the agreed 'Contaminant Chemicals Groups' March 2022' spreadsheet. Two versions of 'contaminant name' are listed:

- 'Contaminant name' reported by the article cited, and
- 'Contaminant synonym' used by ECOTOX or others, if available and different from 'contaminant name'.

Species name

The name of the species studied as reported in the original article. Relevant synonyms, based on WoRMS, are used in the report text.

Life stage studied

Terms defined in MarLIN glossary

- Adult
- Juvenile
- Larvae
- Embryo
- Egg
- Sporophyte
- Gametophyte
- Multiple

Exposure concentration

The experimental concentrations the samples were exposed to, where available, and expressed in reported units and µg/l where possible.

Exposure type

Definitions of the type or route of exposure to the contaminant, adapted from ECOTOX.

Term	Definition
Environmental	Field and incidental exposures, includes via the water column or sediment
Environmental (sediment)	Optional where sediment concentration are paramount (e.g. sedimentary communities)
Flow-through	Continuous or frequent flow through test chamber with no recycling
Food	Introduced via food
Lentic	Static water without measurable flow e.g. lakes, ponds, lagoons
Pulse	Intermittent or fluctuating dosing
Renewal	Without continuous flow of solution, but with occasional renewal of test solutions after prolonged periods, e.g., 24 hours
Spill	Incidental spills
Static	Toxicity tests with aquatic organisms in which no flow of test solution occurs; solutions may remain unchanged throughout the duration of the test.
Tidal	Affected by tides

Study duration

The length of the study and reported by article in hours, days, months or years etc.

Exposure Duration (ECOTOX definition)

The Exposure Duration is the time of actual exposure to the chemical and is expressed as ‘days’. In cases where the observation time is the only duration reported, it is assumed that the Exposure Duration is equivalent to the longest observation time (field: Observed Duration).

For most field studies the ‘Exposure’ and ‘Study Duration’ are identical because it is difficult to determine when the exposure ends. For lab studies the ‘Exposure’ and ‘Study Duration’ may be different, such as when effect measurements were reported from a post-exposure period. For lab

studies with injection, topical, or dietary (e.g. intraperitoneally or by gavage) exposure, 'Exposure and Study Duration' are typically the same.

For a fluctuating or intermittent dosing experiment, the total exposure time is recorded. In some instances, a biological, or qualitative, time is used, such as an exposure time reported as "until hatch", "growing season" or "after the nth egg has been laid".

Effect group (definitions from ECOTOX)

Term	Definition
Accumulation	Measurements and endpoints that characterize the process by which chemicals are taken into and stored in plants or animals; includes lethal body burden
Behaviour/Avoidance,	Activity of an organism represented by three effect groups - avoidance, general behaviour, and feeding behaviour
Biochemical (inc. enzyme(s), hormone(s))	Measurement of biotransformation or metabolism of chemical compounds, modes of toxic action, and biochemical responses in plants and animals; includes three effect groups - biochemical, enzyme and hormone effects
Cellular/ Histology/ Genetic	Measurements and endpoints regarding changes in structure and chemical composition of cells and tissues of plants or animals as related to their functions; includes three effect groups -cellular, genetic and histological effects
Ecosystem process	Measurements and endpoints to track the effects of toxicants on ecosystem processes; includes microbial processes
Growth/ Development/ Morphology	Category encompasses measures of weight and length, and includes effects on development, growth, and morphology
Mortality	Measurements and endpoints where the cause of death is by direct action of the chemical
Multiple	Measurements related to multiple or undefined effect.
No Effect	The author reported an end point but not a specific effect
Physiology/ Immunological/ Injury/ Intoxication	Measurements and endpoints regarding basic activity in cells and tissues of plants or animals; includes four effect groups - injury, immunity, intoxication and general physiological response
Population	Measurements and endpoints relating to a group of organisms or plants of the same species occupying the same area at a given time
Reproduction	Measurements and endpoints to track the effect of toxicants on the reproductive cycle; includes behavioural and physiological measurements

Effect measurement

A description of the effect measured. These are likely to vary between different taxonomic groups. The ECOTOX database includes many more categories than listed below for some of the 'effect groups'; the numbers are given in brackets. Examples of standard 'effect measurement' terms, organized by 'effect group', include:

- Accumulation
  - Body burden

- BCF
- Behaviour/Avoidance
  - Chemical avoidance
  - Substratum avoidance
- Biochemical (ECOTOX =1,641 entries)
  - Acyl-CoA oxidase activity
  - Acetylcholinesterase (AchE) activity
  - Acid phosphatase
  - Catalase (CAT)
  - Cytochrome P450 activity
  - Gamma-Glutamyl Transpeptidase
  - Glutathione disulphide
  - Glutathione peroxidase (GPX),
  - Glutathione reductase (GR),
  - Heat shock proteins
  - Lactate dehydrogenase
  - Lipid peroxidation,
  - Metallothioniens
  - MFO (BPH, CYP-dependent monoxygenase)
  - Multixenotoxicity resistance
  - NADPH-Neo tetrazolium Reductase activity
  - NF-E2-related factor 2 (Nrf2),
  - Superoxide dismutase (SOD)
- Cellular (ECOTOX has 143 entries)
  - DNA damage/Micronuclei/Adduct formation
  - Genotoxicity
  - Haemocyte counts population
  - Phagocytosis
  - Lysosomal membrane stability
  - Ovarian and spermatic follicles
  - Transmembrane sodium energy gradient
  - Transcriptomics
- Ecosystem processes
  - General
  - Reduced/Increased productivity (primary/secondary)
  - Community
- Growth/Development/Morphology
  - Abnormal development/larvae
  - Growth rate
  - Leaf/shoot/rhizome/root elongation
  - Leaf shape/morphology
- Mortality (adult/larval)
  - Adult survival
  - Larval survival
- Physiology/Immunological/Injury/Intoxication
  - Byssal thread production
  - Clearance/filtration rate
  - Excretion rate

    - Larval swimming velocity/ability
    - Respiration rate
    - Condition indices
    - Photosynthetic efficiency
    - PSII function/damage
    - Scope for growth (SFG)
    - Valve gape
- Population
    - Abundance/biomass
    - Condition
    - Cover/canopy
    - Distribution/extent
    - Diversity
    - Population decline (general)
- Reproduction
    - Fecundity
    - Gametogenesis reduction
    - Gonad index
    - Fertilization success/failure
    - Recruitment success
    - Settlement
    - Sexual maturity (rate/age)
    - Sex ratios
    - Imposex

Response site

The part (or type) of the organism where the effect (response) is measured (or observed). ECOTOX has 594 entries, which vary between taxonomic groups. We should expect to add terms as we tackle more taxonomic groups but use ECOTOX definitions where possible. For example:

- Community
- Digestive gland
- Embryo
- Gametes (oocytes and sperm)
- Gonad
- Haemocytes
- Larva
- Leaf/shoot
- Lysosomes
- Muscle tissue
- Rhizomes/roots
- Population
- Seedling
- Soft tissues
- Whole organism (assumes adult)

End points

List of observed end points reported by the articles examined, used for consistency with ECOTOX data, but also includes population level effects due to environmental exposure, spills etc. For example:

- BCFD - Bioconcentration factor calculated using dry weight tissue concentration
- EC_{XX}– Effect concentration at XX percentile
- IC_{XX} - Inhibition concentration at XX percentile
- ID_{XX} - Inhibition dose at XX percentile
- LC_{XX}– Lethal concentration at XX percentile
- LD_{XX} – Lethal dose at XX percentile
- LT_{XX} – Lethal time at XX percentile
- LOEC/L – Lowest Observable-Effect-Concentration/Level: lowest dose (concentration) producing effects that were significantly different (as reported by authors) from responses of controls (LOEAL/LOEC)
- NOEC/L – No Observable-Effect-Concentration/Level: highest dose (concentration) producing effects not significantly different from responses of controls according to author's reported statistical test (NOEAL/NOEC)
- Mortality (e.g. after spills)
- NR-LETH – 100% Mortality
- NR-ZERO – 0% Mortality
- Population loss
- Population decline
- Recruitment failure

Endpoint concentrations

ECOTOX provides a single concentration or range (with or without confidence intervals) for each Endpoint. ECOTOX lists the confidence intervals as a range (min, max). In the 'Evidence summary' different End point concentrations (or ranges) are listed separately. Lethal (100%) is included where papers give a concentration resulting in 100% mortality, which is one endpoint recorded by ECOTOX.

Concentrations are expressed as mg/l (ECOTOX) and/or µg/l.

Mortality (%) reported

The percentage mortality reported in the articles examined, where available.

Ranked mortality

The mortality reported in the articles examined is 'ranked' according to the MarESA resistance scale. For example:

Ranked mortality	Resistance
Severe (>75%)	None
Significant (25-75%)	Low
Some (<25%)	Medium
None (reported)	High
Sublethal	High
Unspecified	Unspecified

Unspecified = mortality is reported but not quantified or no detail provided

Quality/Applicability of Evidence – based on MarESA scales

Summary of evidence

The relevant evidence from the articles is summarized in narrative form, using the standard MarESA format description of evidence.

'Worst-case' mortality

The reported 'end points' and evidence from each article is expressed as a 'worst-case' ranked mortality for each contaminant examined in each article. For example, where the specimens are exposed to a range of concentrations of one chemical and several 'end points' (e.g. EC_{50}, LC_{50}) determined, the 'worst-case' or greatest mortality is reported.

Please note, some papers examined several different combinations of contaminant type and seagrass species. Therefore, the 'worst case' mortality is recorded for each unique species vs. contaminant combination within each paper but not for every experimental permutation. For example, if a paper studied three metals and one herbicide, then we would report the four 'worst case' mortalities rather than every mortality or effect from every concentration tested. However, if the papers examined the same combination on three different species (e.g. in seagrasses) then we would record twelve separate 'worst-case' mortalities.

1.8 Synthesis and communication

The aim of the study is a REA to inform a sensitivity assessment of each of the contaminant pressures against each habitat or the species indicative of sensitivity within each habitat. The resultant sensitivity assessment(s) is presented below. However, the REA approach allows us to qualify the evidence-base as a whole.

The key points from the REA are summarized in report format (see Sections 6&7 below) based on the summary narratives and analysis of the collated evidence. The detailed 'Evidence summaries' are provided in the attached spreadsheets. Only evidence relevant to the 'review question' (the effects of contaminants) on the taxon or habitat interest was recorded in the attached 'Evidence summary'. The evidence is separated into the pressure categories, 'Hydrocarbons and PAHS', 'Transitional metals (inc. organometals)', and 'Synthetics compounds (inc. pesticides, antifoulants, and pharmaceuticals)' and the 'Introduction of other chemicals'.

The results will be disseminated via the MarLIN/MarESA sensitivity assessment web pages.

1.9 Bibliography

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