Sensitivity Assessment of Contaminant Pressures - Selected Amphipods – Evidence review.

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# 1 Introduction

An evidence review of the effects of contaminants on selected species of amphipod was undertaken between December 2024 and March 2025. The evidence review followed the Rapid Evidence Assessment (REA) protocol developed previously (Tyler-Walters *et al.*, 2022).

The resultant 'Amphipod Evidence Summary' spreadsheet (available here) and 'evidence review' that follows benefited from improvements and resultant minor adjustments. The 'evidence summary' template was updated to improve data entry. The improvements included:

- the addition of both the reported and standardised values for the exposure concentrations of contaminants used (where available),
- the addition of both the reported and standardised values for the observed or effect concentrations of contaminants (where available), and
- use of 'common' or 'short' names for chemicals derived from the PubChem<sup>1</sup> database where possible, and
- a standard 'summary narrative' writing style was adopted for consistency in reporting.

In addition, 'contaminant type' is recorded as the function of the chemical (e.g., herbicide, analgesic), rather than the structure of the chemical (e.g. organohalogen, organophosphate), if the information allows.

## 2 Evidence review overview

The literature review focused *Ampelisca* sp., *Bathyporeia* sp., and *Corophium* sp. but also recovered articles<sup>2</sup> on other amphipod genera. Freshwater studies were excluded to keep the literature review manageable.

#### 2.1 Literature review

The initial searches (December 2025) resulted in ca 3,475 hits of which 483 were duplicates (Table 2.1) using the standard search strings developed previously (Tyler-Walters *et al.,* 2022). Only the Web of Science (WoS) science citation index and the ECOTOX<sup>3</sup> Knowledgebase were used and freshwater studies were excluded due to time constraints. The resultant references were screened for relevance based on the proposed REA protocol. Only articles written in English or with readily available English translations were included.

Review stage	No. articles identified/retained	No. articles rejected/removed
Web of Science	3,274	
ECOTOX database	199	
Duplicates removed	3,473	483
MarLIN Endnote additions	3,271	33
Screening	143	3,128
Taken forward**	143	
Not accessible	18	

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Table 2.1	Results of lit	erature	review to	r selected	amphipods
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\*\* Does not include further articles identified from the articles reviewed, or alternative sources

However, the initial literature review returned more articles than could be processed in the time available. Hence, relevant articles on other amphipod genera such as *Hyalella*, *Hyale*, *Orchestia, Echinogammarus, Eohaustorius, Talitrus*, and especially *Gammarus* sp. (with 347



<sup>&</sup>lt;sup>2</sup> The term 'article(s)' or 'study' are used for peer reviewed papers, reports and other publications relevant to the review.

relevant articles alone) were excluded. Furthermore, only articles relevant to hydrocarbons were included for *Ampelisca* sp. However, recent synonyms of *Corophium* (e.g. *Monocorophium*) were included. Screening against the exclusion criteria reduced this number to 143 articles, which were taken forward for detailed review. However, 18 articles could not be accessed.

A number of amphipod genera (e.g. *Ampelisca, Corophium, Eohaustorius, Gammarus, Hyalella* and *Rhepoxynius*), are used as standard toxicity bioassays, especially in contaminated sediment bioassays (Reish, 1993; Chapman & Wang, 2001). Hence, many of the articles reviewed used relevant amphipods as bioassays for the toxicity of sediments contaminated by oil spills, hydrocarbons, PAHs, metals, PCBs and other contaminants or mixtures of multiple contaminant types.

The results and evidence from studies of contaminated sediment are given in the relevant sections below, where the articles both characterize (measure and document) the contaminants within the sediment tested and document a relationship between one contaminant type and the resultant toxicity. However, if the contaminants are not documented, or where a mixture of contaminant types are present and no relationship between one contaminant type and the resultant toxicity is given, the relevant evidence is presented in a separate 'contaminated sediment' section. These contaminant types because contaminants may act synergistically or antagonistically in mixtures and it was not obvious which one or more the contaminants caused the reported toxicity.

Nevertheless, many contaminated sediment studies used cadmium or other metal as a 'reference toxicant' to demonstrate the suitability of the amphipod specimens used for the bioassay. Therefore, the results of cadmium studies are included under the relevant 'transitional metals' section below.

#### 2.2 Overview of results

The articles reviewed reported 'worst-case' ranked mortality ('severe' to 'some') in 81% of results, no mortality ('none') in 4.3% of results, and sublethal effects in 11.4% of results<sup>4</sup>. The level of mortality or sublethal effect was 'unspecified' in the remaining 3.3% of results (Figure

<sup>&</sup>lt;sup>4</sup> The term 'results' refers to the 'worst-case' ranked mortalities listed in the evidence summary spreadsheet, unless otherwise specified.

# 2.1). The detailed evidence is presented in the 'evidence summary spreadsheet' associated with this review.



Figure 2.1. Count of ranked worst-case mortalities due to exposure to contaminants in selected amphipods. Mortality is ranked as follows: 'Severe' (>75%), 'Significant' (25-75%), 'Some' (<25%), 'None' (no mortality reported), and 'Sublethal' effects.



Most articles examined (45%) studied the effects of transitional metals on the amphipods. However, this number is skewed because most of the bioassay studies used inorganic cadmium (usually CdCl<sub>2</sub>) as a reference toxicant in their experiments. The next most studied contaminant types were 'petrochemical hydrocarbons' (inc. oil spills) (15.6%) and 'pesticides/biocides' (15.4%). Amphipods are used to examine the toxicity of contaminant sediment. In this review, 7.6% of the results were derived from contaminated sediment bioassays.



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## 3 Hydrocarbons and PAHs

A total of 89 worst-case mortality results were obtained from 36 articles that studied the effect of hydrocarbons, PAHs and dispersants on amphipods, of which 32.5% examined the effects of oil spills and 22.5% examined the effects of complex hydrocarbons such as crude or fuel oils and/or their water accommodated or saturated fractions (WAF/WSF) (Figure 3.1). The majority (65.5%) of the hydrocarbon and PAHs results were based on studies of *Corophium* spp., followed by *Ampelisca* spp. which provided 28.7% of the results.

## 3.1 Oil spills

The effect of oil spills were reported by 14 articles. The evidence is summarized below.

Amoco Cadiz oil spill - 16th March 1978. Cabioch et al. (1978) examined the effect of the Amoco Cadiz oil spill on sublittoral benthic communities in northern Brittany, particularly the sensitive burrowing Ampelisca amphipod species, while also assessing the impact on other species, such as echinoderms. The study documented the rapid penetration of oil into seabed sediments, leading to severe declines in *Ampelisca* populations, which initially accounted for 90% of the amphipod community. Ampelisca densities dropped from over 5,500 individuals/m<sup>2</sup> to near zero within weeks after the spill, with Ampelisca sarsi being the only species temporarily persisting at reduced numbers (367 individuals/m<sup>2</sup> in early April, declining to 15 individuals/m<sup>2</sup> by late April). Other Ampelisca species, including Ampelisca tenuicornis, Ampelisca brevicornis, and Ampelisca spinipes were eliminated from the affected sediments. The overall number of amphipod species declined from 24 to just seven during this period. In contrast, mollusc and polychaete populations appeared less affected, and Abra alba exhibited substantial recruitment following the disturbance. Cabioch et al. (1978) concluded that Ampelisca species were particularly vulnerable due to their high population densities in fine sands and their dependence on stable, well-oxygenated sediment habitats. The effects on *Ampelisca* populations and their recovery varied depending on hydrodynamic conditions, with finer sediments acting as long-term contamination reservoirs, potentially delaying recolonization, and ecosystem recovery.

*Amoco Cadiz* oil spill – 16<sup>th</sup> March 1978. Dauvin (1987) examined the effect of the *Amoco Cadiz* oil spill on benthic amphipod populations, in particular *Ampelisca sarsi, Ampelisca tenuicornis, Bathyporeia elegans,* and *Corophium crassicorne,* in a long-term study conducted in the Pierre Noire area of the Bay of Morlaix, France.





Figure 3.1. Count of ranked worst-case mortalities due to exposure to 'Hydrocarbons and PAHs' in selected amphipods. Mortality is ranked as follows: 'Severe' (>75%), 'Significant' (25-75%), 'Some' (<25%), 'None (reported)', and 'Sublethal' effects.

Dauvin (1987) monitored amphipod populations from 1978 to 1986, assessing mortality, recolonization, and population recovery following oil contamination. *Ampelisca sarsi* and *Ampelisca tenuicornis* were the most severely impacted, with *Ampelisca* populations



suffering a 99% decline, and five out of six species disappearing completely. *Ampelisca sarsi* initially showed 99.4% population reduction, and while *Ampelisca spinipes* reappeared within one year, *Ampelisca tenuicornis* was not observed again until 1985, nearly eight years later, with total *Ampelisca* densities still 90% lower than pre-spill levels. *Bathyporeia elegans* also experienced severe declines, nearly disappearing from contaminated sediments. However, due to its greater mobility, recolonization occurred within three to five years and populations recovered faster than *Ampelisca* species. *Corophium crassicorne*, a burrowing species, was eliminated from affected areas and was not observed again by the end of the study, indicating the habitat was unsuitable after the spill. Other benthic species, such as polychaetes and the bivalve *Abra alba*, showed higher resilience, with *Abra alba* recovering within two to three years. Dauvin (1987) concluded that *Ampelisca, Bathyporeia*, and *Corophium* species were highly vulnerable to oil pollution due to their sediment-dwelling habits and limited dispersal abilities.

*Amoco Cadiz* oil spill – 16<sup>th</sup> March 1978. Dauvin (1998) examined the effect of the *Amoco Cadiz* oil spill on benthic invertebrate communities, with a focus on *Ampelisca sp.* but also *Bathyporeia sp.*, in a long-term study conducted in the English Channel along the northern coast of France. The study analysed the impact of oil contamination on amphipod populations and compared recovery patterns with other benthic species, including *Abra alba* and polychaete worms. *Ampelisca* populations were severely affected, with some areas reporting complete disappearance in heavily contaminated sediments. Recovery was slow, with significant recolonization not occurring until five to seven years post-spill, due to the persistence of hydrocarbons in sediments. Other benthic species, such as *Abra alba*, showed greater resilience, with populations recovering within two to three years. Dauvin (1998) concluded that *Ampelisca* species were highly sensitive to oil pollution, making them valuable bioindicators of long-term ecological damage, with populations recovering 15 years after the initial spill. However, he noted that recovery rates varied depending on sediment type, hydrocarbon degradation, and larval dispersal dynamics, which influenced the duration for full ecosystem restoration.

*Amoco Cadiz* oil spill – 16<sup>th</sup> March 1978. Gesteira & Dauvin (2000) examined the impact of the *Amoco Cadiz* (1978) and *Aegean Sea* (1992) oil spills on *Ampelisca* populations in softbottom macrobenthic communities in the Bay of Morlaix (western English Channel) and Ría de Ares and Betanzos (northwestern Iberian Peninsula). Both spills led to the near-total disappearance of *Ampelisca*, with minimal recolonization over four years, likely due to their high sensitivity to hydrocarbons (up to 3,152 ppm in sediments), low fecundity, and limited



dispersal ability. Recovery was slow but progressive, whereas polychaete populations remained stable or even increased, suggesting stronger resistance and differing sensitivities among taxa. Gesteira & Dauvin (2000) concluded that *Ampelisca* is highly susceptible to oil pollution. They also proposed that *Ampelisca* could be used as a bioindicator of oil pollution, and they suggested a polychaete/amphipod ratio to assess long-term ecological changes. They emphasized the need for detailed community assessments post-spill but acknowledged site-specific variability in species responses.

Amoco Cadiz oil spill - 16<sup>th</sup> March 1978. Poggiale & Dauvin (2001) examined the effect of the 'Amoco Cadiz' oil spill on the population dynamics of three Ampelisca species Ampelisca armoricana, Ampelisca sarsi, and Ampelisca tenuicornis in the Bay of Morlaix (western English Channel) through a 20-year field study conducted from 1977 to 1996. The study tracked the disappearance and subsequent recolonization of these species following the spill. Hydrocarbon concentrations in sediments peaked at 200 ppm in 1978–1979, declining to below 50 ppm after 1981. The spill led to the near-total disappearance of Ampelisca populations in 1978, with only a few Ampelisca sarsi (<100 individuals per m<sup>2</sup>), persisting in low densities. Recolonization was delayed by the species' demography, lack of pelagic larvae, and habitat isolation. Ampelisca armoricana began recolonization in 1981 but remained at low density until 1987, while Ampelisca sarsi actively recolonized in 1987, and Ampelisca tenuicornis (the most affected species) did not return until 1988. By 1993, Ampelisca densities had reached pre-spill levels (>40,000 ind. m<sup>2</sup>). Poggiale & Dauvin concluded that pollution levels initially dictated recolonization success, followed by competition and environmental conditions. The study highlights the resilience of Ampelisca populations but acknowledges the complexity of recolonization processes and the need for further research into dispersal mechanisms and their difficulty to estimate due to variations with time such as seasons and years.

**Braer oil spill – 5<sup>th</sup> January 1993**. Kingston *et al.* (1997) examined the effects of the *Braer oil* spill on crustacean populations, in particular amphipods, off the coast of Shetland, Scotland. Amphipods, in particular Ampeliscids are very susceptible to oil pollution, such as reported after the *Amoco Cadiz* spill by Cabioch *et al.* (1980). By 1993 Amphipoda were completely absent from areas which saw the largest amount of oil contamination in West Burra, and all stations in the main path of the oil spill had reduced numbers. However, there was no quantified data was reported.



**Deepwater Horizon oil spill – 20<sup>th</sup> April 2010.** Van Eenennaam *et al.* (2018) simulated the effects of the Marine Oil Snow Sedimentation and Flocculent Accumulation (MOSSFA) event that occurred after the *Deepwater Horizon* (DWH) oil spill. The MOSSFA transported ca 14% of the total released oil to the seabed. Laboratory sediment mesocosms were treated with oiled (Macondo oil) sediment or oiled artificial marine snow at a concentration of 10 g/m<sup>2</sup> for 16 days. The controls lacked oil. Selected marine benthos, including *Corophium volutator*, were added after one day. Their behaviour was monitored and their mortality assessed after 16 days. The survival of *Corophium* was significantly reduced in the oil and 'snow and oil' treatments compared to controls. *Corophium* escaped the low oxygen levels at the sediment water interface caused by the addition of artificial marine snow. Artificial snow alone

water interface caused by the addition of artificial marine show. Artificial show alone decreased survival (to 69%) but was not significantly different from controls. The oil treatment severely reduced survival to 9% of controls (91% mortality) while the 'snow and oil' treatment reduced survival to 20% (80% mortality), probably because *Corophium* escaped the snow layer, as above, and possibly fed on the artificial snow (Van Eenennaam *et al.*, 2018). However, the presence of artificial marine snow reduced the biodegradation of oil. Van Eenennaam *et al.* (2018) noted that their study used a high concentration of oil, comparable to the higher exposures experienced during the DWH spill but that organisms would be exposed for longer than 16 days in the field.

*Exxon Valdez* oil spill – 24<sup>th</sup> March 1989. Wolfe (1996) examined the effect of the *Exxon Valdez* oil spill on *Corophium* amphipods, as well as the bioaccumulation of hydrocarbons in Pacific oysters (*Crassostrea gigas*), in a study conducted along the oil-impacted shorelines of Prince William Sound, Alaska. Sediment-dwelling *Corophium* populations were severely affected in heavily oiled intertidal areas, with sharp declines observed as 12.1 to 53.35% mortality after the spill due to direct oil toxicity and habitat contamination. Recovery of *Corophium* populations was slow, as residual oil in sediments continued to impact benthic communities. Wolfe (1996) concluded that the *Exxon Valdez* spill had long-lasting effects on sediment-dwelling invertebrates, with *Corophium* being particularly vulnerable due to its direct exposure to contaminated sediments. However, he noted that natural degradation processes, sediment transport, and bioturbation influenced oil persistence, affecting the process of ecological recovery in varying habitats.

*Hebei Spirit* oil spill – 7<sup>th</sup> December 2007. Lee *et al.* (2013) monitored the toxicity of intertidal sediments for five years after the *Hebei Spirit* oil spill in December 2007, in Taean, Korea. Sediment hydrocarbon contamination was estimated and 10-day sediment toxicity bioassays undertaken using *Monocorophium* (syn. *Corophium*) *uenoi.* The sediments from



14/15 sample sites were significantly toxic to amphipods within four months after the spill. The lowest mortality was 10% but rose to >70% at the most contaminated site. However, most sites were not significantly toxic after eight months, except for the most contaminated site where amphipod mortality was 47%. In 2009, samples from the three most contaminated areas were analysed for 16 PAHs, and sediment toxicity examined. Of the 63 samples taken, 39 were significantly toxic to amphipods, five years after the spill. Amphipod mortality ranged from 14 to 95%, from 11 to 95% and from 11 to 90% across the three sites. There was little change in toxicity with season, however, the number of samples toxic to amphipods decreased from 61% in 2009 to 58% in 2012. The total PAH concentrations (TPAH) that were toxic to amphipods ranged from 1,200 to 291,000 ng/g, 9,150 to 530,000 ng/g and from 2,940 to 72,700 ng/g across the three sites. The lowest TPAH that was toxic to Monocorophium (syn. Corophium) uenoi was 1,200 ng/g. Lee et al. (2013) suggested that 1,200 ng/g was the threshold concentration in amphipods below which no-toxicity was expected. They also reported 10-day LC10 of 2,840 ng/g TPAH and a 10-day LC50 of 36,000 ng/g TPAH together with Effect Range (ER) and Threshold Effect Levels (TEL) for TPAH in Monocorophium (syn. Corophium) uenoi. They noted that their LC50 was three-times higher than obtained for Rhepoxynius abronius after the Exxon Valdez, probably due to that species' higher sensitivity. They concluded that sediment toxicity was reduced rapidly (eight months the spill) in areas subject to clean up but remained in sediment at one site used to store oil from the clean-up operation temporarily.

*Rose Bay* oil spill – 12 May 1990. Roddie *et al.* (1994) examined the toxicity of sediments oiled by the *Rose Bay* oil spill in the Erme estuary, southwest England in 1990. *Corophium volutator* was exposed to sediment samples collected from five oiled sites, together with a negative control (from Poole Harbour) and a positive (polluted) control (from Devonport, Plymouth. The negative control was unexpectedly contaminated by heavy metals. Nevertheless, the sample most contaminated by hydrocarbons (356  $\mu$ g/g wwt) resulted in the significant higher mortality (ca 50% based on Figure 2) in *Corophium* than any of the other samples or controls examined.

**Sea Empress oil spill – 15th February 1996**. Rostron (1998) examined the effects of the Sea Empress oil spill in the Milford Haven Waterway, Pembrokeshire, in 1996 on infauna committees, in particular amphipod populations including *Ampelisca*, *Bathyporeia*, and *Corophium*. Rostron (1998) concluded that amphipod species, especially *Ampelisca* were highly sensitive to oil pollution leading to mortalities amongst populations, compared to other benthic communities such as polychaetes which were less sensitive to oil. Rostron (1998)



cited data from Chasse & Morvan (1978) of different infauna species following the *Amoco Cadiz* spill. After the *Amoco Cadiz* spill *Ampelisca* brevicornis had the lowest survival rate at 0%. Both *Bathyporeia pilosa* and *Bathyporeia sarsi* had survival rates of 10%, and *Corophium volutator* had the highest resistance to the oil pollution with 20% survival reported.

Sea Empress oil spill – 15<sup>th</sup> February 1996. Nikitik & Robinson (2003) examined the impact of the Sea Empress (1996) oil spill on Ampelisca, Corophium, and Bathyporeia populations in the Milford Haven Waterway (Wales). They compared populations pre spill (1993) to post spill (2000). The spill caused a sharp decline in amphipod populations, particularly Ampelisca, while polychaete populations increased, reflecting shifts in community structure. Recovery was evident at all survey sites by 1998, with Ampelisca reaching pre-spill levels in some areas by 2000, though recovery remained incomplete in the middle Haven. Nikitik & Robinson concluded like other oil spill papers that amphipods (Ampelisca in particular) are highly sensitive to oil contamination, making them effective indicators of ecosystem disturbance following oil spills. The authors highlighted the polychaete/amphipod ratio as a potential bioindicator for oil pollution impacts and suggested that other amphipod groups, such as Harpinia spp. and Isaeidae, could be valuable indicators in future assessments. Nikitik & Robinson overall emphasized the need for long-term monitoring to capture sitespecific variability in amphipod responses to oil spills.

*Torrey Canyon* oil spill – 18<sup>th</sup> March 1967. Smith (1968) examined the effect of the *Torrey Canyon* oil spill on marine invertebrates, including *Corophium* amphipods, in intertidal and shallow subtidal habitats along the affected coastline. The study was conducted in oil-contaminated areas of southwest England, assessing the impact of both crude oil and the potential effects of chemical detergents in the sands of the mouth of the river on benthic communities through observations and sediment sampling. *Corophium* populations experienced a decline in heavily contaminated sites, likely due to the direct toxicity of oil, smothering effects on sediment, coupled with any effects from detergents.

*World Prodigy* oil spill – 23<sup>rd</sup> March 1989. Widbom & Oviatt (1994) examined the effect of the '*World Prodigy*' oil spill on macrobenthic crustacean populations in Narragansett Bay, Rhode Island, across five stations with varying degrees of oil exposure, including a control site. The study tracked population changes over five weeks following the spill. At the most heavily impacted station (station 2), where sediment contained 23 µg oil/g dry weight, total amphipod abundance was dominated by *Ampelisca verrilli*, which declined by 86% within two weeks and 93% within five weeks. Significant declines in *Ampelisca abdita* populations at



69% were observed at another station with lower initial oil sedimentation. No significant decline was observed in *Corophium* amphipods. The study confirmed the high sensitivity of *Ampelisca* to oil pollution, consistent with prior field and experimental research. Widbom & Oviatt (1994) suggested that sediment contamination and oil-associated particulates contributed to the mortality observed. They also concluded that amphipods particularly *Ampelisca*, are highly sensitive to oil contamination, especially juvenile specimens. The *World Prodigy* oil spill occurred just after breeding season. Therefore, under 2 mm juveniles were frequently encountered. However, some amphipods may have actively migrated rather than perished, and long-term sublethal effects on reproduction could extend population impacts beyond the study period.

#### 3.2 Petroleum hydrocarbons – oils and dispersed oils

Fourteen of the articles examined the effects of petroleum oils (e.g. crude oil and fuel/bunker oils), and dispersed oils, and are summarized below.

Bonsdorff et al. (1990) examined the effect of water-soluble fractions (WAF) of Ekofisk crude oil on the recruitment of zoobenthos, focusing on Corophium bonelli, a dominant colonizer found in subtidal soft-bottom habitats in Raunefjorden, off Bergen, Norway. The study used field experiments with colonization trays (both covered and uncovered) and larger enclosures at 8 m depth. Oil exposure was conducted in two 14-day periods, with mean concentrations of 30-40 µg/I WAF immediately after injection with WAF. Corophium bonelli showed significant population declines in oil-exposed trays compared to controls, with peak densities of 14 and 36 individuals per 0.01 m<sup>2</sup> in oil and control conditions, respectively. Juvenile survival appeared particularly affected, possibly due to oil-induced embryonic mortality or increased escape responses. However, Bonsdorff et al. (1990) stated that the level of crude oil tested was not particularly lethal to adults. Therefore, they were less effected despite previous papers recordings of high sensitivity to oil pollution in *Corophium* species. Bonsdorff et al. (1990) concluded that even low oil concentrations could negatively impact Corophium bonelli recruitment, although predation in uncovered trays may have masked some effects. These findings align with previous studies suggesting high amphipod sensitivity to oil pollution.

**Briggs et al. (2003)** examined the effect of contaminated sediments on *Corophium volutator* in a laboratory toxicity test using sediments from the NW Hutton oil installation (North Sea) and copper-spiked sediments from Breydon Water (Norfolk, UK). Sediments were tested for toxicity by measuring the turbidity caused *Corophium volutator* activity over a 10-day period.



Copper concentrations ranged from 5 to 1,065 mg/g, and total hydrocarbons from 35 to 1,721 mg/g. The 10-day LC50 was determined as 129 mg/g for copper and 231 mg/g for hydrocarbons. Turbidity at 24 hours correlated with contaminant levels and mortality, suggesting that increased turbidity is a behavioural response to sediment toxicity. Briggs *et al.* (2003) concluded that turbidity measurements could provide a rapid method for assessing sediment toxicity before mortality occurs. However, they noted that sediment grain size could also influence the results. Therefore, Briggs *et al.* (2003) suggested the need for further research in the future to refine the method and assess its applicability across different toxicants

**Brils et al. (2002)** examined the effect of oil contaminated sediment on *Corophium volutator* by exposing them to freshly spiked marine sediment with varying concentrations of Gulf distillate marine grade A (DMA) gas oil and Gulf high viscosity grade 46 (HV46) hydraulic oil in laboratory conditions in the Netherlands. *Corophium volutator* was exposed to DMA and HV46 for 10 days in sediment containing different concentrations of oil, with mortality as the endpoint. The estimated LC50 values were 100 mg/kg dry weight for DMA and 9,138 mg/kg dry weight for HV46, indicating that *Corophium volutator* was significantly more sensitive to DMA exposure. Brils *et al.* (2002) concluded that toxicity was strongly correlated with the lower boiling-point fractions of the oil, particularly within the C10–C19 range. However, aging experiments showed that the toxicity of oil-spiked sediment decreased over time, suggesting that natural weathering processes could reduce the environmental risk of oil contamination.

**Cappello et al. (2015)** examined the effects of oil-spiked sediment on *Corophium orientale* and tested the effects of different bioremediation strategies. Under laboratory conditions, sediment collected from Messina harbour was spiked with 11 g/kg crude oil (Kashagan Fresh Oil, ENI Technology, Italy) and added to five different mesocosms. Each mesocosm was supplied with a continuous flow of seawater from Messina Harbour and was subjected to five bioremediation treatments for 30 days, including only polluted sediment system (SED) and bioremediation systems involving the addition of air (SED+A), slow-release fertiliser (SED+A+F), oil sorbents (SED+A+F+S) and temperature regulation (SED+A+F+T). Over the 30 days, oil degradation was monitored. The physical and chemical variables of the collected sediment were also measured. Sediment samples were collected from each mesocosm at regular intervals (on days 0, 5, 10, 15, 20, 25 and 30) and exposed to juvenile and young adult *Corophium orientale* for 10 days in glass flasks. Each flask contained filtered seawater and 100 randomly selected individuals. All experiments were conducted twice. Mortality was recorded after 10 days. Results found toxicity was highest in the SED-only system at day 0



and day 30. *Corophium orientale* mortality was close to 50% in the sediment samples from SED+A, SED+A+F and SED+A+F+S. Toxicity was low for sediment samples from the control mesocosms (10% mortality) and SED+A+F+T (30% mortality). In the SED+A+F+T mesocosm, approximately 70% of the hydrocarbons were degraded and was, therefore, the best bioremediation system to reduce amphipod mortality. Cappello *et al.* (2015) concluded that the combined bioremediation strategies significantly enhanced hydrocarbon degradation, therefore reducing sediment toxicity and improving the survival rates of *Corophium orientale*.

**Guerra-Garcia et al. (2003)** studied the effects of aliphatic hydrocarbon pollution on softbottom macrobenthic assemblages in Ceuta Harbour, North Africa. The Ceuta Harbour is contaminated by shipping activities and sewage disposal. Sediment grab samples were collected from 21 stations; 15 inside the harbour and six outside the harbour. Macrofauna from the sediments were sorted, identified to species level, and counted. The aliphatic and aromatic hydrocarbons and unresolved complex mixture (an indicator of oil pollution inputs) in the sediment samples were analysed. The chemical analysis identified total aliphatic hydrocarbon concentrations in the sediment varied from 400 to 6,021 ppm, with aliphatic hydrocarbons being more abundant than aromatic ones. Stations inside the harbour were more contaminated by hydrocarbons than stations outside the harbour. Results found a total of 217 macrofaunal species in sediment samples, with *Corophium runcicorne* and *Corophium sextonae* found exclusively in the sediment samples from stations inside the harbour.

**Kienle & Gerhardt (2008)** examined the effects of water-accommodated fractions (WAF) of weathered Forties crude oil on the survival and behaviour of adult *Corophium volutator* collected from Avon estuary, South Devon, in laboratory-based aqueous and sediment exposures. The study used a Multispecies Freshwater Biomonitor, which is an online biomonitor that records behaviour patterns, to analyse locomotion and ventilation behaviour. The Forties crude oil used to prepare the WAFs consisted of mainly paraffines, naphthene and aromatics. In the aqueous exposures, the *Corophium volutator* was exposed to control (0% WAF), 100% WAF and dilutions of 25% and 50% WAF in seawater for two hours. There were six individuals in each treatment. Kienle & Gerhardt (2008) observed hyperactivity (increased swimming activity) with an increase in ventilation in *Corophium volutator* when exposed to 25% and 50% WAF. Kienle & Gerhardt (2008) noted that hyperactivity is an avoidance behaviour and a common toxic effect, observed as a first sign of stress. This was a significant behavioural response. However, narcosis (hypoactivity) was observed in *Corophium volutator* in aqueous exposure to 100% WAF in aqueous experiments, as individuals were lying on the bottom for most of the exposure period. This was not



significantly different from the control, potentially due to high variation in the behaviours of *Corophium volutator*.

In sediment exposures, *Corophium volutator* was exposed to sediment spiked with 100% WAF for two hours. There were six individuals in each treatment. Kienle & Gerhardt (2008) reported that *Corophium volutator* avoided burrowing and re-emergence from the sediment. One individual did not burrow at all, while the other five stayed in the sediment for a certain amount of time. They also reported that the amphipod had a tendency towards hyperactivity but the differences were not significantly different from the control. *Corophium volutator* was observed to be swimming, whereas in the control four out of the six individuals were constantly burrowing. An additional, stress and recovery 130-minute pulse experiment was conducted, exposing six *Corophium volutator* individuals to 50% WAF and seawater for two hours, as described in the aqueous exposure experiment. After two hours, half the solution was removed and replaced with filtered seawater. After another 1.5 hours, all of the solution an increase in hyperactivity (locomotor activity) during both the 130-minute exposure and after the recovery period.

Kienle & Gerhardt (2008) concluded that the effects of WAF on the locomotor activity of *Corophium volutator* was more pronounced in aqueous exposures than sediment exposures. This difference in sensitivity may be due to lower environmental relevance as the amphipods spends most of its time in the sediment. This suggested *Corophium volutator* seeks refuge in sediment. *Corophium volutator* was able to recover after 18 hours but further, longer experiments were needed to prove this conclusion (Kienle & Gerhardt, 2008).

**McLusky (1982)** reviewed the effect of petrochemical effluent on benthic invertebrate communities in the intertidal mudflats of the Kinneil area, Forth Estuary, Scotland, over a six-year period (1975–1980), in particular *Corophium volutator*, while also assessing species such as *Macoma balthica, Hediste diversicolor*, and oligochaete worms. *Corophium volutator* exhibited extreme sensitivity to pollution, with populations absent in areas within 500 meters of effluent discharge and only beginning to reappear at distances greater than 1.5 km from the pollution source. McLusky (1982) concluded that petrochemical pollution had severe impacts on sediment-dwelling amphipods, particularly. *Corophium volutator*, which serves as a sensitive bioindicator of estuarine contamination. However, he noted that recovery potential is influenced by sediment conditions, hydrodynamics, and the persistence of chemical



pollutants, with recolonization occurring more rapidly in areas where pollution sources were removed.

McLusky & Martins (1998) investigated the long-term effects of petrochemical discharge on the faunal composition of an estuarine mudflat over a 20-year period. The study location was the Kinneil intertidal area, in the middle reaches of the Forth estuary, eastern Scotland, which has been subject to the effects of industrial discharges, principally from petrochemical industries (oil refinery and chemical works) since the 1920s. The intertidal fauna in the estuary had been studied annually since 1976, providing over 20 years of data from 90 stations in the estuary. During the study period, the discharges into the estuary had been reduced through a combination of plant closure and the installation of effluent treatment works. In addition, the River Avon that flows across the area had experienced substantial improvements in water quality. Corophium volutator showed a significant increase in abundance after 1985 (Figure 5), which coincided with the closure of the Phenol plant in 1985. Regression analysis indicated that the increase in *Corophium* abundance was correlated significantly with a decrease in chemical effluents from the phenol or acrylonitrile plants and the resultant decrease in BOD (biological oxygen demand). McLusky & Martins (1998) also noted that the effluents at the site were toxic to Corophium, where the 96-hour LC50s were 17.5% (of effluent mixed with seawater) for chemical effluents in 1986, 16% for refinery effluent in 1986 and 90% for refinery effluent in 1995 (cited from Smith, 1987; McLusky & Colbourne, 1995).

**Percy (1977)** studied the effects of crude oil-contaminated sediment on the behavioural response of benthic crustaceans, including *Corophium clarencense*, using a two-choice test. Sediments were spiked with 0.05 ml (described as light), 0.5 ml (medium), 1.0 ml (heavy), and 2.0 ml (heavy+) of four different oil types; Norman Wells, Pembina, Atkinson Point or Venezuela (Tijuana light) crude oil. In laboratory conditions, 60 *Corophium clarencense* individuals were placed in the centre of an exposure chamber containing clean seawater and an alternating pattern of one concentration of oil-spiked sediment and clean sediment on Petri plates arranged around the periphery of the chamber for 1 or 2 hours. At the end of the exposure, the number of individuals in clean and contaminated sediment was counted. The study found that *Corophium clarencense* was neutral when exposed to oil in sediment. There was an indication of consistent avoidance behaviour by *Corophium clarencense* in exposure to crude oil from Atkinson Point but observed distributions in sediment were not statistically significant.



**Sanz-Lázaro & Marín (2009)** examined the effects of oil contamination on amphipods (*Corophium multisetosum* and *Microdeutopus gryllotalpa*)in the laboratory and the field. The amphipods were exposed to sediment spiked with 15 and 60 ml/kg of Maya crude oil for 10 days in static containers in the laboratory and in PVC tubes in the field (Rio de Aveiro, NW Portugal for *Corophium*). *Corophium* mortality increased significantly with increasing oil concentration in both laboratory and field experiments. For example, survival was ca 10% (based on Figure 2) in the high (60 ml/kg oil) treatments in the field and the laboratory. In addition, the macrofaunal community was significantly different in the high (60 ml/kg oil) exposures than the control in Atlantic sites. The abundance of *Corophium* was significantly lower in the oiled treatments compared to controls.

Scarlett et al. (2007) examined the effects of oil contamination on Corophium volutator in both acute and chronic toxicity tests. Cadmium chloride (CdCl<sub>2</sub>) was used as a reference toxicant. Corophium was exposed to 0 to 14 mg/I CdCl<sub>2</sub> for 72 hours. The resultant 72-hour LC50 was 7.45 mg/l CdCl<sub>2</sub>, within the range of 2.7 to 9.9 mg/l reported by Ciarelli et al. (1997). In the acute test, adult *Corophium* were exposed to sediment spiked with nominal concentrations of Alaskan North slope crude oil (at 110, 220, 440 and 880 µg/g), Corexit 9527 (400 mg/l), 100% Water Accommodated Fraction (WAF) and 100% Corexit Dispersed WAF (DWAF) for 10 days. No significant mortality was recorded below 440 µg/g or at 100% WAF. Twenty percent mortality occurred with 100% DWAF but was not significantly different from 25% mortality under Corexit exposure. Twenty five percent mortality occurred at 880 µg/g oil exposure. In the chronic test, neonates (larval) Corophium were exposed sediment spiked to the same concentrations of oil, WAF, dispersant and DWAF as above (except 880  $\mu q/q$ ) for 28 days. Growth rates and reproduction were measured. Mortality was only significantly different to controls in the DWAF treatment (43% mortality). Scarlett et al. (2007) suggested that the oil was more biological available in chemically dispersed exposure. Growth rates were significantly reduced in 110 µg/g and 440 µg/g oil, while 440 µg/g oil had the lowest growth rate of all exposures tested. Growth rates in DWAF were lower than Corexit and WAF treatments. Reproduction occurred in all treatments. However, 440 µg/g oil, DWAF and two replicates of 220  $\mu$ g/g oil had no offspring. Reproduction was significantly less in the DWAF and all oil treatments than in controls, Corexit and WAF treatments. Scarlett et al. (2007) concluded that sediment that was not toxic after the 10-day acute test but could still cause population-level effects on amphipods.

**Suchanek (1993)** reviewed the effect of oil pollution from both chronic and catastrophic spills on various marine benthic invertebrate taxa, including crustaceans, molluscs, echinoderms,



and polychaetes. The study reviewed multiple oil spill events and experimental findings from various locations across the globe, analysing mortality, sublethal effects, and long-term community impacts. Amphipods, particularly ampeliscid species, were identified as highly sensitive to oil contamination, often experiencing severe population declines and failing to recover for at least five years post-spill, likely due to the longevity of oil in sediments. Other crustaceans, such as copepods and crabs, exhibited variable sensitivity, with some populations recovering more quickly. Suchanek (1993) concluded that oil exposure disrupts invertebrate communities through direct toxicity, reduced recruitment, and altered trophic interactions, leading to shifts in species composition. However, he noted that environmental factors such as sediment type, wave action, latitude, season, and suspended sediment can influence oil persistence and alter invertebrate responses to contamination.

#### 3.3 Dispersants

The effects of dispersants themselves were examined by Scarlett *et al.*, 2005, 2007. Scarlett *et al.*, (2005) is summarized below and Scarlett *et al.* (2007) above (Section 3.2).

Scarlett *et al.* (2005) examined the effects of the dispersants Corexit 9527 and Superdispersant-25 (SD-25) on a selection of invertebrates including *Corophium volutator*. Adult *Corophium* were exposed to sediment spiked with 0, 50, 125, 175, 213, 250, 375, and 500 ppm of Corexit 9527 or SD-25 for 48 hours. Behaviour was monitored during the experiment, and survivors transferred to clean water for 24 hours to monitor recovery. No activity was observed after 18 hours at 125 ppm and individuals were stressed at 175 ppm or above. Moribund individuals were observed after 24 hours at 375 and 500 ppm of both dispersants, and after 42 hours at 175 ppm or above. Mortality was 100% at 500 ppm of both dispersants after 48 hours, and at 375 ppm in Corexit 9527. The 48-hour LC50s were 159 ppm for Corexit 9527 and 260 ppm for SD-25, while the NOEC was 125 ppm and LOEC was 175 ppm for both. The mortality to exposure to Corexit 9527 was significantly greater than SD-25 and recovery rates of survivors from SD-25 was higher than form Corexit 9527. Scarlett *et al.* (2005) noted that dispersant concentrations of ≥175 ppm reduced burrowing in the laboratory, which, in the wild, might allow *Corophium* to leave contaminated sediments.

## 3.4 Polyaromatic hydrocarbons (PAHs)

The effects of polyaromatic hydrocarbons (PAHs) of pyrogenic origin were examined by nine articles. The evidence is summarized below, except for Gesteira & Dauvin (2000) which is summarised above (Section 3.1).



Boese et al. (1997) examined the photoinduced toxicity of fluoranthene on seven marine benthic crustaceans, including five amphipod species (Rhepoxynius abronius, Eohaustorius estuarius, Leptocheirus plumulosus, Grandidierella japonica, and Corophium insidiosum), collected from shallow intertidal areas, under controlled laboratory conditions. Specimens were exposed to fluoranthene (1 to 200 µg/l) for 4-day in water-only to assess mortality (LC50) and burial ability (EC50). The exposure was followed by 1-hour UV exposure to evaluate photoinduced effects. In Corophium insidiosum, fluoranthene exposure alone resulted in a 96-hour LC50 of 85 µg/l, which decreased to 32 µg/l after UV exposure, while the EC50 for reburial declined from 54 µg/L to 20 µg/l, indicating an increase in toxicity after UV exposure. Rhepoxynius abronius and Echaustorius estuarius exhibited the highest sensitivity, with LC50s decreasing up to ten-fold after UV exposure, whereas species more naturally exposed to sunlight (Excirclana vancouverensis and Emerita analoga) showed very small to no phototoxic response. A separate experiment exposed the same species to cadmium (CdCl<sub>2</sub>) in a 96-hour toxicity test to compare its effects with fluoranthene, showing no photoinduced toxicity except in Rhepoxynius abronius, where reburial EC50 decreased after UV exposure. Boese et al. (1997) concluded that photoactivation significantly enhances fluoranthene toxicity and that standard toxicity tests conducted under artificial lighting may underestimate environmental risks and emphasized the need for ecologically relevant exposure conditions in sediment toxicity assessments to best simulate conditions occurring in nature. Boese et al. (1997) also noted the relatively short 4-day exposure and 1-hour UV treatment may underestimate the full extent of toxicity that could occur in natural settings over longer periods, as well as species highly sensitive in the study, such as Rhepoxynius abronius, are unlikely to experience direct sunlight exposure in nature, potentially leading to an overestimation of environmental risks.

**Ciarelli** *et al.* **(1999)** examined the effect of sediment bioturbation by the estuarine amphipod *Corophium volutator* on fluoranthene resuspension and its transfer to the filter-feeding blue mussel *Mytilus edulis* in a laboratory-based study conducted in southwestern Netherlands. Experiments were conducted using sediment spiked with fluoranthene at a concentration of 50 mg/g dry weight, with amphipods introduced at different densities (ranging from 100 to 320 individuals per aquarium) and over exposure periods of 10 to 30 days. The study measured total suspended solids (TSS), particulate organic carbon (POC), and fluoranthene concentrations in sediment, pore water, overlying water, and organisms. *Corophium volutator* significantly increased TSS and POC in overlying water, leading to an increased total in aqueous fluoranthene concentrations, which rose from 2.4 mg/l in controls to 4.1 mg/l at low



amphipod density and 5.45 mg/l at high density after 10 days. Amphipods accumulated fluoranthene, with concentrations ranging from 86.5 to 97.8 mg/g dry weight after 10 days but decreasing to 4.7 mg/g by day 30, suggesting depletion over time. Ciarelli *et al.* (1999) concluded that bioturbation enhances the resuspension of sediment-bound contaminants, increasing their bioavailability to filter feeders like *Mytilus edulis*, which accumulated higher fluoranthene concentrations with increasing amphipod density and exposure duration. However, they noted that despite increased fluoranthene levels in the overlying water, the sediment and pore water concentrations stayed mostly the same, suggesting that bioturbation transports contaminants without changing how they are distributed between sediment and water. The authors also acknowledged that while bioturbation can increase contaminant fluxes, natural factors such as water currents and organic matter content may modify these effects in real-world conditions.

**Engraff et al. (2011)** examined the effect of polycyclic aromatic hydrocarbons (PAHs) on the benthic amphipod *Corophium volutator* through passive dosing experiments in marine sediment environments at Ganavan Bay, Oban, Scotland, and Roskilde Fjord, Denmark. The study exposed *Corophium volutator* to single PAHs and mixtures at their aqueous solubility levels under controlled laboratory conditions at 10°C and 21°C for up to 15 days. The lethal concentration for 50% of the population (LC50) was not explicitly reported. However, mortality increased with PAH chemical activity, with an effective chemical activity causing 50% lethality (EA-50) estimated at 0.036 for *Orchomenella pinguis*, another species studied. The study found that PAH mixtures had additive toxicity effects, with some delayed lethality, indicating that PAH metabolites might contribute to toxicity. Engraff *et al.* (2011) concluded that chemical activity could be a useful metric for assessing PAH mixture toxicity in environmental risk management. However, Engraff *et al.* (2011) suggested that further research should focus on confirming how suitable the chemicals are for predicting mixture toxicity across species and conducting longer-duration experiments to capture delayed toxic effects.

**Fisher** *et al.* (2011) examined the effect of polycyclic aromatic hydrocarbons (PAHs) on the marine amphipod *Corophium volutator* using a 10-day sediment bioassay in a laboratory setting conducted in the UK. The amphipods were exposed to twelve low molecular weight PAHs at varying concentrations, with sediment toxicity assessed based on mortality rates. The study determined acute toxicity values (10-day LC50) for each PAH, which ranged from 24 mg/kg for 4-methyldibenzothiophene to over 1,000 mg/kg for anthracene. Phenanthrene was used as a reference compound, and toxic equivalency factors (TEFs) were calculated to



express the relative toxicity of each PAH. Fisher *et al.* (2011) concluded that PAH contamination in sediments can pose a significant risk to benthic organisms, with certain compounds demonstrating much higher toxicity than others. They emphasized the need for further validation of TEFs and extension of the approach to additional PAH compounds. The study acknowledged limitations such as the potential for degradation of PAHs in sediment and the complexity of PAH mixtures in environmental settings.

Spehar et al. (1999) examined the effect of fluoranthene toxicity under fluorescent and ultraviolet (UV) light on a range of freshwater and saltwater species, including the amphipod Ampelisca abdita, in a laboratory-based study conducted at the U.S. Environmental Protection Agency's Atlantic Ecology Division in Narragansett, Rhode Island. Acute 96-hour toxicity tests were performed using fluoranthene concentrations ranging from below detection limits to levels near its solubility threshold in seawater, with separate tests conducted under fluorescent and UV light to evaluate phototoxic effects. In Ampelisca abdita, fluoranthene was toxic under fluorescent light, with a 96-hour LC50 of 67 µg/l (59 to76 µg/l confidence interval), but under UV exposure, toxicity increased significantly, leading to complete mortality at concentrations that were too low to determine an LC50. Other crustaceans, such as the mysid Mysidopsis bahia (LC50 = 31 µg/l under fluorescent light, 1.4 µg/l under UV) and grass shrimp Palaemonetes sp. (LC50 = 142  $\mu$ g/l under fluorescent light, 22  $\mu$ g/l under UV), exhibited similar UV-enhanced toxicity, with LC50 values decreasing by up to 30-fold under UV exposure. Spehar et al. (1999) concluded that fluoranthene toxicity is substantially increased by UV exposure, posing a greater environmental risk than indicated by standard tests under artificial lighting. However, they noted that laboratory conditions may not fully represent natural UV exposure levels, coupled with that UV light could be reduced by dissolved organic matter and suspended particulates in natural waters, leading to reduced phototoxic effects in some habitats.

**Swartz** *et al.* (1990) examined the effect of fluoranthene toxicity in sediment on the marine benthic amphipod *Corophium spinicorne*, alongside *Rhepoxynius abronius*, to evaluate the equilibrium partitioning approach for sediment quality criteria. The study was conducted in a laboratory at the U.S. Environmental Protection Agency Laboratory in Newport, Oregon, using sediment from Yaquina Bay. *Corophium spinicorne*, an epibenthic, tube-dwelling amphipod, was exposed in 10-day sediment toxicity tests at six different fluoranthene concentrations (0 to13.6 mg/kg dry weight) in sediments with three levels of organic carbon (0.18%, 0.31%, and 0.48%) to assess mortality (LC50). Results showed that *Corophium spinicorne* was less sensitive to fluoranthene exposure than *Rhepoxynius abronius*, with an



interstitial water LC50 of 37.9 µg/l compared to 23.8 µg/l for *Rhepoxynius abronius*. The lower sensitivity of *Corophium spinicorne* may be due to its exposure routes, as it constructs U-shaped tubes that allow interaction with overlying water, whereas *Rhepoxynius abronius* is a free-burrowing species exposed directly to interstitial water. Swartz *et al.* (1990) concluded that sediment quality criteria derived from equilibrium partitioning and water quality guidelines are protective of sensitive benthic invertebrates. However, they noted that the study focused on only one chemical, two species, and sandy sediment with low organic carbon, meaning results should not be broadly extrapolated to other species, contaminants, or sediment types.

Werner & Nagel (1997) examined the effects of cadmium, diazinon, dieldrin, and fluoranthene on heat shock protein (HSP) expression in three amphipod species: Hyalella azteca, Ampelisca abdita, and Rhepoxynius abronius. Ampelisca abdita was collected from San Francisco Bay, California, and heat shock protein levels (HSP60 and HSP70) were analysed using western blotting techniques. In Ampelisca abdita, heat shock proteins were primarily detected in the pellet fraction, with significant induction at higher contaminant concentrations. Cadmium at 0.5–2.5 mg/l and diazinon at 30 mg/l significantly increased HSP64 and HSP54 levels, while fluoranthene at 35 mg/l also induced these proteins. However, dieldrin did not induce stress proteins at environmentally relevant concentrations, and higher doses (60 mg/l) inhibited HSP54 expression. The 24-hour LC50 values for Ampelisca abdita were >2.5 mg/l for cadmium, 21 mg/l for diazinon, >60 mg/l for dieldrin, and >100 mg/l for fluoranthene, which indicated that heat shock protein induction occurred at concentrations below lethal thresholds for most contaminants. The study concluded that Ampelisca abdita exhibited a stress protein response that was less sensitive than Hyalella azteca, with HSP60 being the dominant biomarker, which highlighted interspecies variability in stress responses and the importance of species-specific considerations in ecological risk assessments.

### 3.5 Petroleum hydrocarbons – others

Brown *et al.* (1999) examined the effects of 4-nonylphenol on *Corophium* sp. Nonylphenol is used to manufacture antioxidants, emulsifiers, detergents, and lubricating oils. Redmond & Scott (1987) examined the toxicity of phenol in *Ampelisca abdita* (cited from ECOTOX). Krang (2007) examined the effect of naphthalene exposure on mating behaviour in *Corophium volutator* and is summarized below.

Brown et al. (1999) examined the effect of long-term exposure to 4-nonylphenol (NP) on the growth, sexual differentiation, and reproduction of the marine amphipod *Corophium volutator* 



in a laboratory-based study conducted at the University of Plymouth, UK. Amphipods were collected from the intertidal mudflats of the Tamar Estuary, southwest England, and exposed to NP in seawater for up to 120 days at concentrations ranging from 10 to 200 mg/l. A 96hour acute toxicity test determined an LC50 of 1,670 mg/l, while chronic exposures resulted in significant reductions in survival and growth, with a 30-day LC50 of 270 mg/l. Growth was reported to be reduced in individuals as well as fewer reaching reproductive size. Overall populations decline figures reported as 10.9% (50 mg/L), 17.4% (100 mg/L), and 78.3% (200 mg/L). However, fertility increased in exposed females, and males exhibited significantly elongated second antennae at concentrations of 50 mg/l and above (p < 0.001). Brown et al. (1999) hypothesized that NP may act via the androgenic gland, altering male secondary sexual characteristics, potentially leading to a selective disadvantage due to increased predation risk. They concluded that NP exposure affects Corophium volutator populations through altered growth and reproduction, with potential ecological consequences. However, the study acknowledged that nominal NP concentrations may overestimate actual exposure levels due to adsorption to sediment and degradation over time, with further research needed to determine long-term population-level effects in natural environments.

**Krang (2007)** examined the effect of naphthalene exposure on mating behaviour in *Corophium volutator.* Receptive female *Corophium* release pheromones to attract males to their burrows. Krane (2007) used 'Y-maze' experiments, in which males were allowed to choose between two paths, one towards sediment with 50 females and one to sediment with none. Males were exposed to sediment spiked with 0, 0.5, 5, 50 µg/g naphthalene for three days prior to the behavioural 'Y-maze' experiments. Male response to pheromones was significantly reduced by exposure to 0.5 and 5 µg/g naphthalene, their search response was reduced by 27 to 45% and they could no longer find females using their olfactory sense. Krang (2007) noted that the actual sediment concentration of naphthalene was 2 to 20 times lower than the nominal concentration. No significant effects were noted at 50 µg/g naphthalene as the amphipods avoided burrowing in the sediment. Females produced and released pheromones irrespectively of the naphthalene exposure. Krang (2007) concluded that naphthalene contamination could adversely affect reproduction and, hence, the population in contaminated areas.



## 4 Transitional metals and organometals

A total of 173 results (ranked 'worst-case' mortalities) were obtained from 69 articles that examined the effects of transitional metals and organometals on amphipod species. *Corophium* spp. was the most studied species in the articles reviewed, which provided the most results from studies of metals (73.7%) (Figure 4.1).



Figure 4.1. Count of ranked worst-case mortalities due to exposure to 'Transitional metals and organometals' in selected amphipods. Mortality is ranked as follows: 'Severe' (>75%), 'Significant' (25-75%), 'Some' (<25%), 'None (reported)', and 'Sublethal' effects.



## 4.1 Transitional metals

The most studied metals were cadmium (29.5% of results), copper (16.7% of results) and zinc (15.6% of results). However, cadmium (often as CdCl<sub>2</sub>) was often used as a reference toxicant in sediment bioassays (Figure 4.2). The evidence is summarized below.



Figure 4.2. Count of ranked worst-case mortalities due to exposure to separate 'Transitional metals.' Mortality is ranked as follows: 'Severe' (>75%), 'Significant' (25-75%), 'Some' (<25%), 'None (reported)', and 'Sublethal' effects.



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Annicchiarico et al. (2007) examined the effects of heavy metal contamination in marine sediment on Corophium insidiosum and other aquatic species using sediment toxicity bioassays. Sediment samples were collected from four contaminated sites in Mar Piccolo of Taranto and one control uncontaminated site in the Taranto Gulf. Also, organism samples were collected from Mar Piccolo and acclimated in the laboratory prior to the study. The amount of total organic matter and trace metals of mercury (Hg), cadmium (Cd), copper (Cu), lead (Pb) and Nickel (Ni) in sediment samples were analysed, and the sediment was tested with young adult amphipod species. Results showed higher mortality rates of Corophium insidiosum at contaminated sites 1 and 2, where sediment exhibited highest levels of metal contamination, compared to the control site. In contrast, sites 3 and 4, where metal contamination was lower, Corophium insidiosum experienced lower mortality rates that did not significantly differ from the control. In addition, young adults were placed in beakers containing specific dissolved toxicants. The individuals were exposed to one control and five different concentrations of copper, cadmium and mercury ranging from 0.00325 to 12.5 mg/l for 96 hours, to examine the species' sensitivity. Survivors were recorded after 96 hours. The resultant 96-hour LC50 values for Corophium insidiosum were 0.47 mg/l for copper, 1.68 mg/l for cadmium and 0.07 mg/l for mercury. The results suggest Corophium insidiosum has limited tolerance to sediments containing high levels of metal contaminants. The authors concluded that *Corophium insidiosum* was a useful tool for assessing sediment toxicity but noted, Corophium insidiosum has limited contact with sediment as it remains in its tubes which reduced its direct exposure to contaminated sediment.

**Bat (1997)** examined the effects of copper (Cu), zinc (Zn) and cadmium (Cd) on *Corophium volutator* collected from Ythan estuary, Aberdeenshire, Scotland through laboratory-based bioaccumulation bioassays using seawater with uncontaminated sediment and contaminated sediment with uncontaminated seawater. In the first experiment, adult *Corophium volutator* were placed in containers containing seawater contaminated with 0.1, 1.0, 10.0 ppm of Cu, Zn and Cd, using four replicate containers for each concentration, and three controls containing uncontaminated seawater. There were 20 adults in each container. Individuals were exposed for 96 hours, and mortality recorded daily. Results from the first experiment found that 10.0 ppm concentrations of all metals were the most toxic to *Corophium volutator*. At 10.0 ppm of Cu 10% of amphipods died, at 10.0 ppm of Zn 30% of amphipods died, and at 10.0 ppm of Cd 45% of amphipods died after 96 hours of exposure.

In the second experiment, adult *Corophium volutator* were placed in containers containing sediment contaminated with 10, 30, 50 ug/g of Cu and Zn and 5, 10, 30 ug/g of Cd. There



were 20 adults added to each container. Individuals were exposed for 96 hours, and mortality was recorded at 3, 6, 24, 48, 72 and 96 hours. Results from the second experiment found similar results. At 50 ppm of Cu in sediment, 45% of amphipods died, at 50 ppm of Zn 60% of amphipods died and at 30 ppm of Cd 75% of amphipods died after 96 hours of exposure. Overall, all experimental results found survival decreased with increasing Cu, Zn and Cd concentrations, revealing toxic effects of metal concentrations. Cu was less toxic to *Corophium volutator* than Zn or Cd, suggesting the amphipod is more resistant to Cu.

**Bat et al. (1998)** examined the effect of copper (Cu), zinc (Zn) and cadmium (Cd) on the amphipod *Corophium volutator* collected from Ythan estuary, Scotland, through laboratory-based bioaccumulation sediment bioassays and behavioural tests. In the first experiment to examine the toxicity of the metals in seawater, *Corophium volutator* was exposed to 0.1, 1, 10, 100 mg/l of Cu, Zn or Cd in seawater with and without clean uncontaminated sediment; there were eight replicate containers and eight control containers. Each container contained 20 adult *Corophium volutator*, which were exposed for 96 hours, and mortality recorded daily. The survival of *Corophium volutator* decreased with increasing concentrations of Cu, Zn and Cd in seawater. They reported LC50 values of 20.74 mg/l of Cu, 9.79 mg/l of Zn, and 9.03 mg/l of Cd in the absence of sediment, and LC50 values of 37.59 mg/l of Cu, 14.12 mg/l of Zn and 12.50 mg/l with sediment. The presence of sediment reduced the metal toxicity.

The second experiment created choice and no-choice conditions to examine the behaviour responses to contaminated sediment. In both choice and no-choice conditions, a control and 24 containers were used for each metal. In the no-choice conditions, *Corophium volutator* was exposed to clean sediment contaminated with 0, 20, 40, 60, 80, 100 µg/l of Cu, Zn, and Cd for 96 hours. There were 20 adults used in each container, and the survival and burrowing activity of the amphipod was recorded at 3, 6, 24, 48, 72 and 96 hours. In the choice experiment, the containers were divided into quarters, with two opposite quarters filled with clean sediment and the other two quarters filled with sediment contaminated by concentrations of Cu, Zn or Zn as described above for 96 hours. There were 20 amphipods placed in contaminated quarters, the survival was recorded daily, and burrowing behaviour was recorded after 96 hours. In no-choice conditions, the survival of *Corophium volutator* and burrowing activity decreased with increasing metal concentrations. After three hours, more than 50% of amphipods burrowed at concentrations of either 30 ug/g Cu of sediment, 25 ug/g Zn, 10 ug/g Cd or less. In the choice conditions, amphipods avoided burrowing in contaminated sediment, which increased in higher concentrations of metals.



The third experiment examined the separate and combined effects of Cd and Zn contaminated sediments on the bioaccumulation and survivorship of *Corophium volutator*. Clean sediments were contaminated as above with either Cd or Zn or both metals for 96 hours. There were four replicate containers for each concentration, 20 amphipods per container and mortality was recorded after 96 hours. The results found when both metals were present mortality was less than with Cd concentrations alone. At Cd concentrations of 32.7  $\mu$ g/g and 21  $\mu$ g/g, mortality was 70% and 40% respectively, but the presence of zinc significantly decreased the mortality to 43% and 31%, respectively. There was little mortality found in Zn only sediment, as at 91  $\mu$ g/g and 38  $\mu$ g/g, mortality was 12% and 2%, respectively. This suggested that Cd and Zn mixtures were less toxic than just Cd. Overall, results from all experiments revealed metal contamination in sediments is toxic to *Corophium volutator* with Cd and Zn being more toxic than Cu (Bat *et al.*, 1998).

Bat & Raffaelli (1998) examined the effects of metals copper (Cu), zinc (Zn) and cadmium (Cd) on the behaviour and mortality of *Corophium volutator* from the Ythan estuary, Aberdeenshire, through laboratory-based sediment toxicity bioassays. The study spiked intertidal sediment from the Ythan estuary with stock solutions of Cu, Zn or Cd and achieved different metal concentrations through serial dilutions. The Cu sediment concentrations were  $3 \mu g/g$  (control),  $5 \mu g/g$  (control),  $16 \mu g/g$ ,  $25 \mu g/g$ ,  $30 \mu g/g$ ,  $41 \mu g/g$ ,  $57 \mu g/g$ , and  $101 \mu g/g$ . The Zn sediment concentrations were 7  $\mu$ g/g (control), 8  $\mu$ g/g (control), 19  $\mu$ g/g, 21  $\mu$ g/g, 27  $\mu g/g$ , 45  $\mu g/g$ , 59  $\mu g/g$ , and 99  $\mu g/g$ . The Cd sediment concentrations were <0.02  $\mu g/g$ . (control), 0.412 µg/g, 0.548 µg/g, 1.20 µg/g, 9.18 µg/g, 17.67 µg/g, 28.27 µg/g, and 35 µg/g. Twenty adult Corophium volutator individuals were exposed to each concentration of spiked sediment for 10 days, and four replicates per metal concentration were used. The survival, emergence from the sediment (floating on the water surface or lying on top of sediment) and the ability to rebury in the sediment were recorded daily. After 10 days, sediment and seawater samples were analysed for Cu, Zn and Cd, and Corophium volutator individuals were analysed for total Cu, Zn and Cd in their tissues. The concentrations of Cu in the water after 10 days of exposure were <0.03 µg/ml, 0.19 µg/ml, 0.21 µg/ml, and 0.38 µg/ml. The concentrations of Zn in the water after 10 days of exposure were <0.025  $\mu$ g/ml, 0.184  $\mu$ g/ml, 0.266 µg/ml, and 0.430 µg/ml. The concentrations of Cd in the water after 10 days of exposure were <0.02  $\mu$ g/ml, 0.07  $\mu$ g/ml, and 0.11  $\mu$ g/ml.

They reported that *Corophium volutator* mortality increased with increasing Cu, Zn and Cd sediment concentrations. Over 90% of amphipods survived to the end of exposure to sediment concentrations of 25  $\mu$ g/g of Cu and 21  $\mu$ g/g of Zn or less. However, only 70% of



amphipods survived at the end of the exposure to Cd sediment concentrations of 9.18  $\mu$ g/g or less. The 10-day LC50 value of 37  $\mu$ g/g of Cu, 32  $\mu$ g/g of Zn and 14  $\mu$ g/g of Cd. Cadmium was more toxic to *Corophium volutator* than Cu and Zn. There was 100% mortality in the highest concentrations of Cu (101  $\mu$ g/g), Zn (99  $\mu$ g/g), and Cd (35  $\mu$ g/g).

Emergence from the sediment was not significantly different from the control at the highest concentrations as no amphipods emerged after 10 days, as all died during the experiment and no reburial was recorded. Emergence from sediment varied greatly between concentrations of 30 to 57 µg/g of Cu, 26 to 59 µg/g of Zn and 9.18 to 28.27 µg/g of Cd. Emergence from sediment increased with increasing metal concentrations, showing the metal contaminants had sublethal effects. The emergence reduces exposure to the metal in the sediment. Out of the 1,240 surviving individuals in the experiments, 94% (1,160 individuals) were able to rebury at the end of the 10 days. The recorded reburial 10-day EC50 values were 31.66 µg/g of Cu, 28.59 µg/g of Zn and 9.30 µg/g of Cd. Bat & Raffaelli (1998) suggested that *Corophium volutator* can burrow and behave normally once exposure to contamination has ended, after 10 days. Bat & Raffaelli (1998) concluded that *Corophium volutator* was sensitive to metal-contaminated sediment, but Cd is more toxic to this species than Cu or Zn. Evidence presented shows *Corophium volutator* is robust and able to tolerate high levels of some contaminants (Bat & Raffaelli, 1998).

Bell et al. (2020) examined the effect of dissolved galvanic anode-derived metal ion exposure on Corophium volutator, collected from uncontaminated reference sites at Sylt and Norderney, Germany, using laboratory-based acute toxicity tests and metal uptake experiments. Galvanic anodes are typically composed of aluminium-zinc-indium alloys and are used for cathodic corrosion protection of offshore wind turbine support structures. These anodes gradually dissolve in water, releasing aluminium, indium, and zinc ions that contaminate the surrounding environment. In the experiment, Corophium volutator was exposed to dissolved aluminium anode material (nominal concentrations of 1, 10, 100 mg/kg seawater), standard aluminium (nominal concentrations of 1, 10, 100 mg/kg seawater), and standard zinc (0.1, 1, 10 mg/kg seawater) for 10 days with sediment and three days without sediment. There were two to six replicates per concentration and exposure type (with or without sediment). Aluminium anode material had no acute toxicity on *Corophium volutator*. Significant effects were found at the highest concentrations of aluminium (100 mg/kg) and zinc (10 mg/kg). In the aluminium exposures, the highest mortality of Corophium volutator (17.5% mortality in 100 mg/kg treatment) was in the absence of sediment. Statistically significant differences were found between exposure types (with or without sediment) in only


the 100 mg/kg of aluminium treatments. However, in the zinc exposures, the highest mortality (52.5% mortality in the 10 mg/kg treatment) was in the presence of sediment. In addition, in the metal uptake experiments, *Corophium volutator* accumulated metals in proportions similar to the anode's composition. Bell *et al.* (2020) suggested that the presence of sediment appeared to mitigate toxicity. The evidence indicated that there was little direct environmental threat in the use of galvanic anodes on wind turbine structures in the marine environment.

**Berry et al. (1996)** examined the effects of metal contamination on *Ampelisca abdita* collected from the Pettaquamscutt River, Rhode Island, through laboratory-based static renewal water-only and spiked sediment toxicity tests. In the water-only experiment, *Ampelisca abdita* was exposed to five unspecified concentrations of each metal cadmium (Cd), copper (Cu), lead (Pb), nickel (Ni), zinc (Zn) and a control, in glass-containing jars for ten days. There were two replicates per concentration, ten *Ampelisca abdita* in each jar and mortality was recorded daily. The results found 10-day LC50 values of 36 µg/l for Cd, 20.5 µg/l for Cu, 3,020 µg/l for Pb, 2,400 µg/l for Ni and 343 µg/l for Zn. In the spiked sediment tests, *Ampelisca abdita* was exposed to unspecified metal-spiked sediment from Ninigret Pond and Long Island Sound. The authors reported that the bioavailability and toxicity of metals depended on sediment type and ratio of acid-volatile sulphide (AVS) to simultaneously extracted metal (SEM) in the sediment.

**Berry et al. (1999)** examined the effects of silver (as silver nitrate) on *Ampelisca abdita* collected from Pettaquamscutt River, Rhode Island through laboratory-based renewal water-only and spiked sediment toxicity tests. In the water-only experiment, *Ampelisca abdita* was exposed to 6.5  $\mu$ g/l, 11  $\mu$ g/l, 18  $\mu$ g/l, 30  $\mu$ g/l, 50  $\mu$ g/l, 84  $\mu$ g/l and 140  $\mu$ g/l of silver nitrate (nominal concentrations) in glass-containing jars for ten days. In a second test, the measured concentrations of silver nitrate were 3.57  $\mu$ g/l, 7.05  $\mu$ g/l, 12.8  $\mu$ g/l, 22.6  $\mu$ g/l, 39.9  $\mu$ g/l and 76.8  $\mu$ g/l. There were two replicates per concentration, ten *Ampelisca abdita* in each jar and mortality was recorded daily. The experiment was carried out twice. The results from exposure to nominal concentrations found 10-day LC50 values of 44  $\mu$ g/l and 27  $\mu$ g/l of silver nitrate, and the measured 10-day LC50 value was 20  $\mu$ g/l of silver nitrate. The percentage mortality was lowest in low silver concentrations, for example, there was 10% and 35% mortality in *Ampelisca abdita* exposed to 6.5  $\mu$ g/l and 3.57  $\mu$ g/l of silver, respectively. The highest percentage of mortality (80%) was found in higher silver concentrations (140  $\mu$ g/l and 39.9  $\mu$ g/l). In the spiked sediment tests, *Ampelisca abdita* was exposed to unspecified metal-spiked sediment from Ninigret Pond and Long Island Sound. From this, the authors noted



that the bioavailability and toxicity of metals depended on sediment type and ratio of acidvolatile sulphide (AVS) to simultaneously extracted metal (SEM) in the sediment.

Bigongiari et al. (2004) investigated the effects of temperature on the sensitivity of Corophium orientale, collected from Magra River, Liguria, Italy, to reference toxicant cadmium (Cd). In laboratory conditions, Corophium orientale was exposed to control, 0.5 mg/l, 1.0 mg/l, 2.0 mg/l, 4.0 mg/l and 8.0 mg/l of cadmium chloride at different temperature variations (10°C, 15°C, 20°C, 25°C and 30°C) for 96 hours. The experiment was conducted three times, in August, September and January, and new amphipods were collected each month and exposed to the same environmental conditions. The study found that Corophium orientale is very sensitive to variations in temperature. The evidence (from Figure 2; Bigongiari et al., 2004) suggests that Cd is more toxic to the amphipod in higher temperatures, as the calculated 96-hour LC50 values decreased as temperatures increased. Corophium orientale samples in August were more sensitive to Cd exposures, with a constant and low LC50 value at all experimental temperatures. The temperature range 15°C to 20°C is the most suitable for Corophium orientale, at 15°C the 96-hour LC50 value is 4.38 mg/I Cd and at 20°C the 96-hour LC50 value is 1.56 mg/I Cd. Bigongiari et al. (2004) noted that amphipods sampled during the winter period have low metabolic activity, which could reduce their sensitivity to toxicants. This can be seen in the results, in exposure to Cd at low temperatures, there were higher LC50 values recorded. In contrast, it was also noted that elevated metabolic activity could enhance sensitivity of amphipods collected in the summer, with these amphipods having high mortality at higher temperatures above 20°C and higher sensitivity to Cd exposure.

**Briggs et al. (2003)** examined the effect of contaminated sediments on *Corophium volutator* in a laboratory toxicity test using sediments from the NW Hutton oil installation (North Sea) and copper-spiked sediments from Breydon Water (Norfolk, UK). Sediments were tested for toxicity by measuring the turbidity caused *Corophium volutator* activity over a 10-day period. Copper concentrations ranged from 5 to 1,065 mg/g, and total hydrocarbons from 35 to 1,721 mg/g. The 10-day LC50 was determined as 129 mg/g for copper and 231 mg/g for hydrocarbons. Turbidity at 24 hours correlated with contaminant levels and mortality, suggesting that increased turbidity is a behavioural response to sediment toxicity. Briggs *et al.* (2003) concluded that turbidity measurements could provide a rapid method for assessing sediment toxicity before mortality occurs. However, they noted that sediment grain size could also influence the results. Therefore, Briggs *et al.* (2003) suggested the need for further



research in the future to refine the method and assess its applicability across different toxicants.

Bryant et al. (1984) examined the effects of hexavalent chromium (as potassium dichromate) on *Corophium volutator* collected from the Tayport, Eastern Scotland. The study considered the impact of temperature and salinity on the toxicity of chromium to the Corophium volutator. In laboratory conditions, 20 Corophium volutator were exposed to 2, 4, 8, 16, 32, 64 and 128 ppm of chromium at various temperatures (5, 10, 15°C) and salinities (5, 10, 15, 20, 25, 30, 35, 40 ppt) for 384 hours. Mortality was recorded daily and noted as a lack of muscular or pleopod activity in the Corophium volutator. The time until 50% mortality was recorded (LT50) and LC50 values were recorded at 24, 48, 96, 192 and 384 hours. The effects of chromium on *Corophium volutator* depended on both temperature and salinity. Toxicity increased with higher temperature and lower salinity, and the greatest tolerance was at lower temperatures and higher salinities. The authors noted that an increase in salinity (5 to 30 ppt) reduced the impact of chromium, but as chromium concentrations increased, the interaction between salinity and chromium was reduced. Chromium concentrations above 64 ppm caused rapid mortality, suggesting Corophium volutator was highly sensitive to chromium contamination. Values for the LC50 and LT50 values of Corophium volutator for each metal can be seen in the evidence summary spreadsheet.

**Bryant et al. (1985a)** examined the effects of arsenic on *Corophium volutator* collected from Tay estuary, Tayport, Scotland through static acute toxicity tests in laboratory conditions. The study considered the impact of temperature and salinity on the toxicity of arsenic to the *Corophium volutator. Corophium volutator* was exposed to 1, 2, 4, 8, 16, 32, 64 and 128 ppm of arsenic at various temperatures (5, 10, 15°C) and salinities (5, 10, 15, 25, 35 ppt) for 384 hours. In the study, 20 individuals were used for each combination of temperature, salinity, and arsenic; mortality was recorded daily. The time until 50% mortality was recorded (LT50) and LC50 values were recorded at 24, 48, 96, 192 and 384 hours. The results found high arsenic concentrations and high temperatures significantly decreased LT50s, but salinity had little effect on toxicity. The toxicity of arsenic increased with time, as LC50 values were lower at longer exposure times. Values of LC50 and LT50 of *Corophium volutator* for each metal can be seen in the evidence summary spreadsheet. Bryant *et al.* (1985a) highlighted the importance of considering temperature when assessing arsenic toxicity.

**Bryant et al. (1985b)** examined the effects of nickel (Ni) and zinc (Zn) on *Corophium volutator* collected from Tay estuary, Tayport, Scotland, through static toxicity tests in



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laboratory conditions. The study considered the impact of temperature and salinity on the toxicity of Ni and Zn to the Corophium volutator. In the Ni experiment, Corophium volutator was exposed to 2, 4, 8, 16, 32, 64 and 128 ppm of Ni at various temperatures (5, 10, 15°C) and salinities (5, 10, 15, 25, 35 ppt) for 384 hours. In the Zn experiment, Corophium volutator was exposed to 1, 2, 8, 16, 32, 64 and 128 ppm of Zn at various temperatures (5, 10, 15°C) and salinities (5, 10, 15, 25, 35 ppt) for 384 hours. In both Zn and Ni experiments, 20 individuals were used for each combination of temperature, salinity, and metal; mortality was recorded daily. Test solutions were changed daily. The time until 50% mortality was recorded (LT50) and LC50 values were recorded at 24, 48, 96, 192 and 384 hours. The mean survival time of *Corophium volutator* decreased with increasing Ni concentrations, in all temperature and salinity conditions. An increase in salinity increased the survival time of Corophium volutator in all Ni concentrations and all temperatures. This indicated that salinity mitigates Ni toxicity, while salinity reduced survival time. Ni toxicity was higher in low salinity and high temperature conditions. The LC50 values for Ni ranged from 5 to 54 ppm. The median survival time of Corophium volutator decreased with increasing Zn concentrations. Toxicity was higher at lower salinities and higher temperatures. Higher salinities reduced the toxic effects of Zn on Corophium volutator, while higher temperatures increased toxicity. The LC50 values decreased with exposure time, indicating prolonged exposure time increased the risk of mortality. The LC50 values for Zn ranged from 1 to 16 ppm. The authors concluded that the concentration of Zn and Ni, temperature, and salinity all significantly affected the medium survival time of Corophium volutator. Corophium volutator was more sensitive to Ni and Zi than Macoma balthica, another species tested in the study. The full list of results recorded for the LC50 and LT50 values of Corophium volutator for each metal can be seen in the evidence summary spreadsheet.

**Conradi & Depledge (1998)** examined the effects of copper (Cu) concentrations on the life history, growth, and reproduction of *Corophium volutator* from the Tamar Estuary, in laboratory conditions. To examine sublethal effects on growth, 50 male and female *Corophium volutator* juveniles were exposed to a control and 0.2, 0.4, 0.6, 0.8 and 1 mg/l of Cu for 100 days. Mortality was recorded twice a week and growth (measured as cephalic length and total length) was recorded once a week. Copper exposure significantly reduced the lifespan of *Corophium volutator*, especially at higher concentrations. For example, in the control, the average life span of individuals was over 100 days, but individuals exposed to 1 mg/l Cu survived 55 days. Some of the populations exposed to lower concentrations (0.2 and 0.4 mg/l Cu) were able to survive up to 100 days, but the density of surviving individuals was



reduced to 32.4%. Juvenile mortality was highest at 1 mg/l Cu where most individuals died early and did not reach maturity. At 0.4, 0.6 and 0.8 mg/l Cu concentrations, juvenile mortality was low initially but increased as exposure time increased. At 0.2 mg/l Cu there was high juvenile mortality early on (at 36 days). Longevity (time until 50% of the population died) and life expectancy decreased with increasing copper concentrations. The longevity at 0.2 mg/l Cu concentration was 67.5 days, at 0.4 mg/l Cu was 87 days, 0.6 mg/l Cu was 73.5 days, 0.8 mg/l was 57 days, and 1 mg/l Cu was 17.5 days. In addition, Cu significantly reduced the growth rates of *Corophium volutator*, with average body length decreasing as Cu concentrations increased. Individuals exposed to 0.2 mg/l Cu concentrations reached an average length of 7 mm, those exposed to 0.4 mg/l Cu reached an average length of 5.3 mm (males) and 4.4 mm (females), and those exposed to 0.6 mg/l Cu reached an average length of 4.2 mm (males) and 4.0 mm (females). At 0.8 mg/l Cu the growth rate was low with maximum length reached at 4 mm after 77 days. *Corophium volutator* were unable to grow at 1 mg/l Cu.

To examine the effects of Cu on the reproduction of *Corophium volutator*, eight mature females (but no ovigerous females) and four mature males were exposed to the same Cu concentrations as in the growth experiment. The female developmental stage was recorded twice a week, with maturity described as the number of ovigerous females in the population and fertility as the number of newborn individuals per female. Respiration rate was measured in males only. Cu exposure delayed sexual differentiation and delayed maturation in Corophium volutator. It took longer for the amphipods to reach sexual maturity as Cu concentrations increased. For example, Corophium volutator exposed to the control, 0.2 mg/l and 0.4 mg/l of Cu reached sexual maturity at an estimated 58 days, but this was delayed to 77 days at 0.6 mg/l of Cu. Individuals did not reach sexual maturity at 0.8 mg/l of Cu. The length of mature adults was significantly affected by Cu concentrations, as length decreased as Cu concentrations increased in both males and females. After 77 days, there were fewer mature adults. The percentages of mature individuals decreased from 97.3% in the control to 87.5% at 0.2 mg/l, 52.3% at 0.4 mg/l, 41.6% at 0.6 mg/l, and 9.3% at 0.8 mg/l of Cu. Cu concentrations significantly impacted reproduction and fertility, reducing the number of ovigerous females and causing increased offspring mortality. Neonates born in 0.2 mg/l and 0.4 mg/l of Cu could survive until the end of the experiment, but at 0.6 mg/l Cu and above, mortality was high (9.6 to 36%). There was no impact on respiration or mating behaviours, but the study results showed that increasing Cu concentrations caused a reduction in survival and reproductive success. Conradi & Depledge (1998) concluded that despite the tolerance



to high concentrations of copper, in this study, copper had negative effects on the survival, growth and reproduction of *Corophium volutator*.

**Conradi & Depledge (1999)** examined the sublethal effects of zinc (Zn) on *Corophium volutator* from the Tamar Estuary, through growth and reproduction experiments in laboratory conditions.

In the growth experiment, groups of 50 seven-day-old Corophium volutator juveniles were placed in glass tanks containing filtered water and sediment. The amphipod was exposed to less than 0.001 (control), 0.2, 0.4, 0.6 and 0.8 mg/l of Zn for 100 days. There were two replicates per treatment. Mortality was recorded twice a week, and growth (measured as cephalic length and total length) was recorded once a week. They found that Zn exposure reduced the survival rates, longevity, and life expectancy of *Corophium volutator*. Survival decreased with increasing Zn concentrations. In the 0.2 mg/l Zn exposure populations decreased by 31% compared to the control. At 0.4 mg/l Zn, populations decreased by 32%, and at 0.6 mg/I Zn populations decreased by 41%. While there was high juvenile mortality in exposure to 0.2, 0.4, and 0.6 mg/l Zn, the highest concentration of Zn (0.8 mg/l) had significant mortality and juveniles only survived up to 70 days (100% mortality taken from Figure 1). The life expectancy was significantly reduced in higher concentrations. Zn negatively impacted the growth of Corophium volutator, in high Zn concentrations (0.8 mg/l Zn) growth was severely stunted as individuals only reached a maximum length of 4.4 mm, compared to a maximum length of 8 mm in the control group. The specific growth rate also declined with increasing Zn concentrations. At 0.6 and 0.8 mg/l, there was a 9% and 24.6% decrease in specific growth rate, respectively. The length of individuals at the different concentrations were not significantly different but were significantly different from the control population lengths.

In the reproduction experiment, eight mature females (but no ovigerous females) and four mature males were exposed to the same Zn concentrations as in the growth experiment. The female developmental stage was recorded twice a week, with maturity described as the number of ovigerous females in the population and fertility as the number of newborn individuals per female. Conradi & Depledge (1999) reported that Zn exposure significantly impaired *Corophium volutator* reproduction, affecting maturation, fertility, and offspring survival. Sexual differentiation and female maturation were delayed in Zn concentrations. Female survival decreased and females exposed to 0.8 mg/l Zn did not reach sexual maturation. The sex ratio was not significantly affected by Zn exposure. The maturation of



ovigerous females was reduced by Zn. Fertility was also affected, as the maximum number of juveniles observed in populations exposed to 0.2 mg/l Zn was six (Conradi & Depledge, 1999)

**DeWitt** *et al.* (1992) examined the effects of cadmium (as cadmium chloride) on *Ampelisca abdita* from the Pettaquamscutt River, Narragansett, through static laboratory-based toxicity tests. *Ampelisca abdita* was exposed to cadmium concentrations between 0.19 to 6 mg/l for 96 hours in water-only exposures. The amphipod had acute sensitivity to cadmium, with a nominal cadmium concentration 96-hour LC50 value of 1.09 mg/l and an estimated free cadmium ion concentration 96-hour LC50 of 0.07 ppm.

**DeWitt** *et al.* **(1999)** studied the effects of cadmium (Cd) in interstitial (pore) water on amphipod *Chaetocorophium* cf. *lucasi* from Waingaro Landing, New Zealand, in both static laboratory and field conditions. The study focused on the ratio of acid-volatile sulphide (AVS) to simultaneously extracted metal (SEM) in the sediment but did measure the Cd in the interstitial water.

In the laboratory experiment (*in vitro*), Cd spiked sediment was prepared and the measured interstitial water concentrations of Cd were less than 0.001, 0.004, 0.006, 0.54, 9.7, 18, 51 and 118 mg/l. The amphipods were added to test beakers, containing the spiked sediment with overlying seawater that was changed daily for 10 days. In the laboratory, interstitial water Cd concentrations declined by approximately 27% after 10 days. DeWitt *et al.* (1999) reported that the 10-day LC50 value based on interstitial water was 0.41 mg/l of Cd, the NOEC value was 0.023 mg/l Cd and the LOEC was 0.46 mg/l Cd. There was 100% lethality (NR-LETH) recorded at interstitial water Cd concentrations of 8.6 mg/l.

In the field experiments (*in situ*) *Chaetocorophium* cf. *lucasi* was deployed in cages on the mudflat to experience natural tidal flushing, where the measured interstitial water concentrations of Cd were 0.008, 0.004, 0.013, 0.48, 1.3, 9.1, 17 and 41 mg/l. Juvenile *Chaetocorophium* cf. *lucasi* was also found in the cages and was recorded separately. In the field, the natural tidal flushing resulted in a 76% decrease in interstitial water Cd concentrations after 10 days. They found that the 10-day LC50 value based on interstitial water was 1.7 mg/l of Cd and the NOEC value was 0.22 mg/l Cd. The LOEC was 0.65 mg/l Cd, with 37% lethality. There was 75% lethality recorded at an interstitial water Cd concentration of 3.5 mg/l. The sensitivity of juveniles (measured as the number of juveniles counted) was significantly higher than the survival endpoint of adults, suggesting juveniles may be a more sensitive indicator for Cd exposure in sediment toxicity tests. Results for



juveniles found the 10-day LC50 value based on interstitial water was 0.48 mg/l of Cd, the NOEC was 0.22 mg/l Cd, and the LOEC was 0.65 mg/l Cd.

DeWitt *et al.* (1999) indicated that *Chaetocorophium* cf. *lucasi* was more sensitive to Cd in the laboratory compared to the same sediment concentrations in the field. More concentrations caused 100% mortality in the *in vitro* exposure than the *in situ* exposure, suggesting there were higher interstitial Cd concentrations in the *in vitro* exposures. In the static laboratory conditions, Cd accumulated in the overlying water, whereas it was likely dispersed in the field.

In addition, DeWitt *et al.* (1999) conducted a reference toxicant test, using cadmium chloride in a 96-hour water-only exposure test to measure the sensitivity of the *Chaetocorophium* cf. *lucasi* population used in the sediment toxicity tests. *Chaetocorophium* cf. *lucasi* was exposed to serial dilutions of cadmium chloride dissolved in seawater. Results calculated a 96-hour LC50 value of 1.2 mg/l of Cd. This LC50 result was comparable with the 10-day LC50 value from the *in situ* experiment but was significantly higher than the 10-day LC50 value from the *in vitro* experiment. DeWitt *et al.* (1999) concluded that interstitial water Cd is a suitable measure for assessing Cd toxicity, though field conditions may mitigate exposure effects compared to static laboratory tests.

Erdem & Meadows (1980) studied the effects of mercury on the burrowing behaviour of Corophium volutator, through laboratory-based choice and no-choice experiments. Sediments and Corophium volutator were collected from Langbank, Firth of Clyde, Scotland. For the no-choice experiment, individuals were placed in dishes containing sediment which had been treated with varying concentrations of mercury (0.001 ppm, 0.1 ppm, 10 ppm and 1000 ppm) for 21 hours of exposure, and the burrowing behaviour was recorded at 3, 6, 9 and 21 hours. There were two replicates in this test, and the results of these were not significantly different, so they were summed. The percentage of individuals burrowing with time increased in all the mercury-treated sediments. The authors found little difference between the percentage of individuals burrowing (more than 90% burrowed) in the control sediment and the sediment treated with 0.001 ppm, 0.1 ppm and 10 ppm mercury. The results were statistically similar. However, there were no individuals burrowed in the sediment treated with 1000 ppm mercury. In the choice experiment, individuals were placed in dishes that had bottoms divided into four quarters, two opposite quarters contained the control sediment, and the other two contained sediment treated with one of four mercury concentrations (0.001 ppm, 0.1 ppm, 10 ppm and 1000 ppm). This presented a choice for the



amphipod to burrow in either untreated (control) sediment or mercury-treated sediment. *Corophium volutator* was exposed for 21 hours. There were two replicates in this test, and the results of these were not significantly different, so they were summed. More individuals burrowed in the control sediment than in the treated sediment at all mercury concentrations. However, there was no significant difference in the proportions burrowing in the control sediment and 0.001 ppm, 0.1 ppm and 10 ppm mercury-treated sediment. However, more *Corophium volutator* individuals avoided burrowing in 1000 ppm mercury concentrations than in the other treated sediments. Overall, the authors concluded that *Corophium volutator* burrowing was inhibited in sediments treated by 1000 ppm mercury, while individuals avoided sediment treated with the lowest concentration of mercury (0.001 ppm) when given a choice of untreated sediment. Erdem & Meadows (1980) noted that these results are expected as invertebrates often burrow in unsuitable sediments if a suitable one is unavailable. Exposure to mercury influenced the burrowing behaviour of *Corophium volutator*.

**Eriksson & Weeks (1994)** examined the effects of copper (Cu) and oxygen deficiency on *Corophium volutator* in laboratory-based renewal experiments. Specimens were collected from two populations in the Gullmar Fjord, West Sweden: one from the Fiskebackski marina, an enclosed bay at the mouth of the fjord with a high sediment Cu concentration and another from uncontaminated bay, Toresrod, further into the fjord. In the study, groups of sixteen (eleven females and five females) *Corophium volutator* from each population were exposed to control (<0.1 µg/l Cu) and Cu concentrations of 50 µg/l and 100 µg/l for 14 days, both alone and in combination with various degrees of oxygenation. Two replicated series of nine treatments were carried out and measurements of egg production, mortality of females and body copper concentrations (dry weight) were recorded. Exposure to Cu resulted in a significant reduction in egg production in both populations. The authors noted that egg production in individuals from the marina populations was less tolerant to enhanced Cu concentrations, as greater declines in egg production were seen in these individuals compared to the uncontaminated Torserod, despite living in a Cu enriched local environment.

The percentage mortality due to Cu contamination alone (with 100% oxygen saturation) was lower than the percentage mortality caused by decreased oxygen saturation. For example, at 100% oxygen saturation, individuals from the marina experienced 6.3% mortality in 50  $\mu$ g/l Cu exposure and 0% mortality in 100  $\mu$ g/l Cu exposure, and individuals from the uncontaminated site experienced 0% mortality in 50  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure and 5.9% mortality in 100  $\mu$ g/l Cu exposure an



response observed in individuals from both contaminated and uncontaminated sites, there was no significant difference in the mortality between the two populations in all levels of oxygenation. Amphipods under hypoxic conditions experienced behavioural changes in burrowing behaviour and tube building, and therefore, were sensitive to prolonged hypoxia. They also found that Cu concentrations of 50  $\mu$ g/l and 100  $\mu$ g/l significantly increased the body copper concentrations in the amphipods from both populations.

Gaion et al. (2013) examined the toxicity of different arsenic compounds to Corophium orientale from the Magra River, Italy. The study used two arsenic compounds; arsenate, which is an abundant form in sediment and expected to be toxic, and dimethyl-arsenate (DMA), which is considered to be moderately toxic and may be present in contaminated sediment. In laboratory conditions, Corophium orientale was exposed to 1, 5, 10, 20, 25 and 50 ppm nominal concentrations of arsenate and DMA in water, spiked natural sediment and spiked quartz sand for 96 hours. There were three replicates for each concentration and arsenic compound. Overall, the results found that the toxicity measured for arsenate was higher than the toxicity of DMA in all exposures. There were differences in the toxicity in spiked quartz sand, spiked natural sediment and water. Water exposure caused the highest mortalities, especially for arsenate which had an LC50 value of 3.51 mg/l compared to a 96hour LC50 value of 54.65 mg/l for DMA. The lethal concentration curve for arsenate revealed water toxicity was statistically different from both spiked quartz sand and spiked natural sediment, but there was no difference seen between both sediment exposure types. Similar patterns were seen in mortality recorded in the arsenate spiked sediment and spiked quartz sand, as there was a significant difference recorded at 25 ppm arsenate. Toxicity was lower in spiked natural sediment. The recorded LC50 value of arsenate spiked natural sediment was 34.27 mg/l and the LC50 value of arsenate spiked quartz sand was 25.26 mg/l. To contrast, the DMA exposure in natural sediment and quartz sand had the same toxicity, apart from exposures of 50 ppm DMA which had the same toxicity as exposure in water. The recorded LC50 value of DMA spiked natural sediment was 52.19 mg/l and the LC50 value of DMA spiked quartz sand was 827.35 mg/l. The findings demonstrated that arsenic toxicity depended on the chemical form and matrix of exposure (i.e. sediment or water) (Gaion et al., 2013).

**Guerra et al. (2007)** examined the effects of sediment on *Corophium insidiosum* before dredging the inner channel of the Pialassa Baiona Iagoon, Italy. The study used laboratory-based static sediment toxicity tests and a before-after-control-impact (BACI) approach. Sediment samples were collected from 12 sites (impacted and non-impacted). There were six



impacted sites (that will be dredged); three in the channel (described as BAC 1, 2 and 3) and three in an adjacent pond (described as POL 1, POL 2 and VEN 5); and there were six nonimpacted sites; three sites in the channel (described as TBF 1, 3 and 4) and three sites in the pond (described as RIS 1, 2 and 3). Four replicate sample sediments were collected from each site. The metal concentrations of copper, cadmium, chromium, nickel, lead, and mercury in the sediment were determined. The mean measured concentrations of metals in all twelve sediment samples were reported in Table 2, and these were compared to sediment quality guidelines. The copper, chromium and zinc concentrations exceeded guidelines in some sediment samples while the mercury concentrations consistently exceeded thresholds. However, lead and cadmium concentrations in sediment did not exceed guideline thresholds. *Corophium insidiosum* and native sediment (used as a negative control) were collected from uncontaminated Valli di Comacchio Lagoon and exposed to the sediment samples in glass beakers containing overlying seawater for 10 days. Guerra et al. (2007) reported that the survival of Corophium insidiosum in sediment from all sites ranged from 76% to 97%. A significantly lower amphipod survival was seen in sediments from site TBF 4, RIS 2, RIS 3 and POL 3 compared to survival in the native sediment control, suggesting a toxic effect. Guerra et al. (2007) reported survival as high and homogeneous in impacted channel sites (BAC 1, 2 and 3). But in the non-impacted pond sites (RIS 1, 2 and 3), where dredging would not occur, survival tended to be lower. The sediments from non-impacted channels (TBF 1, 3 and 4) and impacted ponds (POL 1, POL 2 and VEN 5) were heterogeneous. However, there were no significant differences found in the survival of Corophium insidiosum between the treatments and sites. Guerra et al. (2007) concluded that the survival of Corophium insidiosum was not significantly correlated with trace metal concentrations in the sediment, so there was no evident spatial pattern in sediment toxicity.

**Haley & Kurnas (1996)** examined the toxicity of dissolved nickel in seawater on *Ampelisca abdita* from Narraganset, Rhode Island, US, through laboratory-based sediment toxicity assays. The study added twenty *Ampelisca abdita* individuals to test chambers containing seawater and unidentified concentrations of ground nickel-coated graphite (NCG) fibres in sediment, for 10 days. After this period, the dissolved nickel concentrations in the overlying water column and mortality were measured. Dissolved nickel concentrations were 0.5 mg/l to 3.1 mg/l. They reported an LC50 value of 40 mg/l for NCG exposure to *Ampelisca abdita*. The authors concluded that NCG fibres were moderately toxic to *Ampelisca abdita*. It was noted that the NCG fibres were mixed in the top 1 cm of the sediment in each chamber,



therefore *Ampelisca abdita* was in direct contact with the contaminant in a closed system, and these concentrations may not reflect realistic environmental exposures.

Hansen *et al.* (1996) examined the chronic effects of cadmium in sediments on the abundance of various marine organisms including amphipods *Corophium acutum, Corophium* sp. and *Ampelisca abdita*. In laboratory conditions, the amphipods were exposed to sediment spiked with nominal cadmium and acid-volatile sulphide ratios (cadmium/AVS) of 0.1, 0.8 and 3.0, which resulted in interstitial cadmium concentrations of 4  $\mu$ g/l (<3 to 10  $\mu$ g/l), 33  $\mu$ g/l (24 to 157  $\mu$ g/l) and 94,000 (28,000 to 174,000  $\mu$ g/l). The species were exposed for 117 days. The results found no significant differences in the number of individuals of the tested amphipod species that colonized each cadmium spiked treatment, even at high exposure concentrations. No effects were observed in all tested organisms exposed to 4  $\mu$ g/l of interstitial cadmium concentrations. However, at higher exposure concentrations, the presence and absence of other species had changed.

Ho et al. (1999b) examined the toxicity of five heavy metals; copper (Cu), cadmium (Cd), lead (Pb), nickel (Ni) and zinc (Zn) to Ampelisca abdita from Narragansett Bay, at three different pHs to characterise pH dependent toxicity. Laboratory-based 48-hour acute static assays were conducted, exposing five Ampelisca abdita to different metal concentrations at pH levels 7, 8 and 9. Exposure concentrations for the tests included, measured metal concentrations 0.614 to 5.100 mg/l of Cd, 0.009 to 0.274 mg/l of Cu and 1.630 to 25.19 mg/l of Pb and stock solutions 1050.30 mg/l of Ni and 1315.6 mg/l of Zn. The measured metal concentrations for Ni and Zn were based on the metal stock solutions as samples were lost during the study. Copper was reported to be the most toxic metal, and the only metal that decreased toxicity with decreasing pH. The IC50 values for Cu were 0.16 mg/l at pH 7, 0.09 mg/l at pH 8 and 0.03 mg/l at pH 9. The toxicity for all other metals remained constant in all pH levels. The IC50 values for Cd were 1.78 mg/l at pH 7, 1.26 mg/l at pH 8 and 1.54 mg/l at pH 9. The IC50 values for Zn were 2.86 mg/l at pH 7, 4.47 mg/l at pH 8 and 1.97 mg/l at pH 9. The IC50 values for Ni were 7.66 mg/l at pH 7, 10.1 mg/l at pH 8 and 9.40 mg/l at pH 9. Lead was the least toxic metal, with IC50 values of 12.3 mg/l at pH 7, 11.3 mg/l at pH 8 and 6.8 mg/l at pH 9. Ho et al. (1999b) found that metal toxicity for Ampelisca abdita was generally dependent on pH levels, and changes in toxicity were metal specific.

**Hong & Reish (1987)** examined the effects of cadmium (Cd) on *Corophium insidiosum* and other marine amphipods from Southern California, through laboratory-based static acute toxicity bioassays. *Corophium insidiosum* was exposed to four unidentified diluted



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concentrations of cadmium from a 1,000 mg/l stock solution of cadmium chloride and a control for seven days. There were three replicates of each concentration and control, and two test amphipods were placed in each container. The results found 96-hour LC50 and 7-day LC50 values of Cd to *Corophium insidiosum* were 1.27 mg/l and 0.51 mg/l, respectively. *Corophium insidiosum* was one of the most tolerant of the seven amphipods tested in the

**Hyne & Everett (1998)** examined the effects of ammonia and copper (Cu) to *Corophium* sp., through laboratory-based static nonrenewal water-only exposures and freshwater sediment toxicity tests. In the water-only toxicity tests, adult *Corophium* sp. collected from the Colo River, Australia, were exposed to five unidentified concentrations of copper chloride and total ammonia and a control for four to ten days. The toxicants were dissolved in freshwater only or a mixture of seawater and freshwater. There were five amphipods in each concentration treatment and five replicates of each treatment. Mortality and salinity were recorded every day. In the laboratory, the collected amphipod adults were cultured in salinities 5 ppt and 10 ppt for 42 days, to recover juvenile *Corophium* sp., which were also used in this toxicity test. The results from juveniles cultured in 5 ppt salinity had a 96-hour LC50 value for copper of 9  $\mu g/l$  and juveniles cultured in 10 ppt had a 96-hour LC50 value for copper of 28.5  $\mu g/l$ . The authors concluded that *Corophium* sp. was sensitive to exposure to copper and ammonia. It was also noted that many juvenile amphipods were observed and collected from 5 ppt and 15 ppt cultures, but only a few were recovered from higher salinity cultures (33 ppt).

**Kater et al. (2000)** investigated the seasonal changes in the acute toxicity of cadmium (Cd) on *Corophium volutator. Corophium volutator* was collected from Oesterput, Eastern Scheldt, The Netherlands, where the seawater temperatures range from an average 5°C in January to 20°C in August. In laboratory conditions, *Corophium volutator* was exposed to 0, 1, 1.8, 3.6, 7.5 and 11 mg/l nominal concentrations of cadmium chloride diluted in natural seawater or artificial seawater for 72 hours. Experiments were conducted on amphipods ten days after collection, and on amphipods that were held in the laboratory for up to five months. The actual concentrations of Cd were measured at the end of the experiment. Each test beaker with Cd spiked concentrations contained twenty randomly selected *Corophium volutator*, and the number of surviving individuals in each beaker was counted after 72 hours. This study was conducted regularly from 1991 to 1998, resulting in 80 experiments in total, to observe seasonal variations. There was significant seasonal variation in the toxicity of cadmium. The lowest sensitivity (highest 72-hour LC50 values) was recorded in the winter period (November to April), and the highest sensitivity (lowest 72-hour LC50 values) was recorded

experiment.



in the summer period (July to September). Results from April had a high LC50 value and were significantly different from other months. Similar results were observed for the newly collected individuals, and the individuals held in the laboratory. There were no significant differences between the LC50 values for *Corophium volutator* exposed to Cd in natural and artificial seawater, nor was there a significant correlation between LC50 values and other measured factors such as organism size or percentage of gravid females. Kater *et al.* (2000) concluded that the observed seasonal variation in the toxicity of Cd to *Corophium volutator* was not caused by the fact that organisms are collected in the field or by the variation in seawater used. LC50 values were reported in their Figure 3 (see Kater *et al.*, 2000) but could not be extracted.

**Khayrallah (1985)** examined the effect of mercuric chloride and ethyl mercuric chloride on *Bathyporeia pilosa*, a sand-dwelling amphipod, through toxicity tests conducted in the Tay Estuary, Scotland. The study used 108 factorial combinations, testing three salinities (10, 20, 35%), three temperatures (0, 10, 20°C), and six mercury concentrations (up to 0.75 mg/l). Toxicity was directly related to concentration and temperature but inversely related to salinity. Mortality increased with higher mercury exposure, especially at lower salinities and higher temperatures. The mean survival time (MST) for *Bathyporeia pilosa* exposed to 0.75 mg/l mercuric chloride at 35% salinity was 57.6 hours, while for ethyl mercuric chloride, it ranged from 6 to 24 hours. The organic mercury compound was more lethal and was absorbed nearly twice as fast as the inorganic form, though lethal concentrations, particularly in estuarine environments with fluctuating salinities and temperatures, could significantly impact *Bathyporeia pilosa* populations, with potential ecological consequences for its predators and the wider ecosystem.

**King et al. (2006)** examined the effects of copper (Cu) and zinc (Zn) on *Corophium colo* from Hawkesbury River, Australia, and other amphipod species, through laboratory-based water only and sediment static acute toxicity tests. In the water only experiments, adult *Corophium colo* were exposed to unspecified concentrations of Cu and Zn dissolved in test water, for 4 days. Survival was recorded at the end of the exposure duration, and LOEC, NOEC and LC50 values were calculated. The results found that Cu and Zn concentrations in the test water decreased due to uptake by amphipods or absorption onto the test glass beakers, but no concentration values were given. *Corophium colo* was less sensitive to dissolved Cu, with an LC50 value and LOEC value of more than 950 µg/l and NOEC value of 950 µg/l. The LC50 value and LOEC value of exposure to Zn were more than 4500 µg/l and the NOEC



value of Zn was 4500. They suggested there were no observed effects of dissolved Zn and Cu on *Corophium colo* in this study. In the whole sediment toxicity tests, *Corophium colo* was exposed to sediment spiked with nominal concentrations 1,300 mg/kg of Cu or 4,000 mg/kg of Zn in beakers with overlying water. Adults were exposed for 10 days and less than 7-dayold juveniles were exposed for 4 days. Survival was recorded at the end of the exposure duration. After exposure to Corophium colo, the final measured Cu concentration in the pore water was 24  $\mu$ g/l and in the overlying water was 56  $\mu$ g/l. The final measured Zn concentration in pore water was 170 µg/l and in the overlying water was 110 µg/l. Exposure to Cu spiked sediments and Zn spiked sediments had little effect on the survival of adult *Corophium colo*, with no significant difference found in the percentage survival. There was around 10% mortality in adult Corophium colo exposed to Cu (based on Figure 1) and around 2% mortality in adult Corophium colo exposed to Zn (based on Figure 2). For all amphipods tested, the juveniles were more sensitive to Cd spiked sediments and Zn spiked sediments than the adults, except for juvenile Corophium colo in which survival was not affected by exposure to Zn. There was around 90% mortality in juvenile Corophium colo exposed to Cu, and around 20% mortality in juvenile *Corophium colo* exposed to Zn (based on figure 3) Corophium colo was the least sensitive amphipod studied.

**Kohn et al. (1994)** studied the effects of total ammonia, un-ionized ammonia, and cadmium (Cd) on amphipods from the USA including *Ampelisca abdita* from San Francisco Bay. In laboratory conditions, the study conducted a range finding test to find estimated LC50 values before conducting definitive toxicity tests, for both ammonia and cadmium exposures. In the range finding experiment, *Ampelisca abdita* was exposed to 0 mg/l, 125 mg/l, 250 mg/l and 500 mg/l nominal concentrations of total ammonia in seawater, for 96 hours, with three replicates for each concentration. The estimated total ammonia LC50 value from range finding test was expected to be <100 mg/l. The definitive toxicity experiments exposed the amphipods to 0 mg/l, 9.375 mg/l, 18.75 mg/l, 37.5 mg/l, 75 mg/l and 150 mg/l nominal concentrations of un-ionized ammonia for 96 hours, with five replicates for each concentration. Amphipods were observed daily, and mortality was recorded after 96 hours of exposure.

The definitive tests for *Ampelisca abdita* were repeated with a new population due to unexpected low survival in the control, as the amphipod appeared to be sensitive to the shipping, acclimation and holding conditions of the study. Initial ammonia results were rejected and the repeat results reported. In the repeat, *Ampelisca abdita* was exposed to total



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ammonia concentrations of 5.9 to 134 mg/l and un-ionized ammonia concentrations of 0.10 to 2.20 mg/l. Results found that Ampelisca abdita was sensitive to ammonia, with a 96-hour LC50 value of 49.8 mg/l for total ammonia and a LC50 value of 0.83 mg/l of un-ionized ammonia. Ampelisca abdita was three times more sensitive to ammonia than Grandidierella japonia, another amphipod tested in the study. The ammonia toxicity tests were water-only with no sediment, but a sediment control treatment was used to compare response of populations with and without sediment. Results found that the survival in water-only exposure of ammonia was not significantly different to the sediment controls, showing that the absence of sediment did not affect amphipod survival in the definitive ammonia tests. In addition, each population used for ammonia testing (this is three populations for *Ampelisca abdita*) was also exposed to up to 4 mg/l of the reference toxicant cadmium in seawater for 96 hours, to assess the health and response of test populations. Results calculated 96-hour LC50 value of 0.63 mg/I Cd for the range finding Ampelisca abdita population, LC50 of 1.32 mg/I Cd for the initial Ampelisca abdita population, and a LC50 of 0.94 mg/l Cd for the repeat Ampelisca abdita population. Ampelisca abdita was around twice as sensitive to Cd as Rhephoxynius abronius, another amphipod species tested in the study. Overall, ammonia was shown to be acutely toxic to sediment-dwelling amphipods, but Ampelisca abdita had the greatest sensitivity out of the four amphipods tested (Kohn et al., 1994).

Lera et al. (2008) examined the effects of cadmium chloride (CdCl<sub>2</sub>) and sodium lauryl sulphate (SLS or sodium dodecyl sulphate SDS) on two populations of Corophium orientale collected monthly from August 2003 to September 2004, from the outfalls of two rivers, Magra and Serchio River, Italy. In laboratory conditions, multiple static water-only tests were conducted. Corophium orientale was exposed to 0, 0.8, 1.6, 3.2, 6.4 and 12.8 mg/l of Cd or 0, 0.625, 1.25, 2.5, 5, 10 and 20 mg/l of SLS for 96 hours. There were two replicates per treatment and 20 individual Corophium orientale per replicate. Survival was recorded at the end of the experiment.

The mortality rate increased as Cd concentration increased in both Corophium orientale populations. The mean 96-hour LC50 for the Corophium orientale population from the Magra River and the Serchio River was 3.71 mg/l and 4.34 mg/l of Cd, respectively. In Cd treatments, both populations observed a similar pattern of increasing 96-hour LC50 values from summer to winter. For the Serchio River population, the 96-hour LC50 went from 1.36 mg/I Cd in August 2003 to 7.23 mg/I Cd in February 2004, and for the Magra River population, the 96-hour LC50 went from 1.21 mg/l Cd in August 2003 to 5.01 mg/l Cd in April 2004. Lera et al. (2008) suggested that this pattern of increasing LC50 value could be related



to the decrease of the temperature in the winter months, with higher LC50 values for both populations found at temperatures lower or equal to 13°C.

The results from the SLS treatments were similar. The mortality rates increased as SLS concentrations increased in both populations. For *Corophium orientale* from the Magra River, the 96-hour LC50 values increased from summer to winter months, like in the Cd treatments. The 96-hour LC50 value was 3.14 mg/I SLS in October 2003 (temperature 17.5°C) and 6.23 mg/I SLS in February 2004 (temperature 8.4°C). Evidence from the Serchio River population was not enough to describe an annual pattern, but there was no significant difference between the two populations.

**Lussier** *et al.* (1998) examined the effects of dissolved metal concentrations of selenium on *Ampelisca abdita* from the Narrow River, Narragansett, Rhode Island, USA, through laboratory-based acute static toxicity tests. *Ampelisca abdita* was exposed to <5  $\mu$ g/l, 456  $\mu$ g/l and 985  $\mu$ g/l measured concentrations of dissolved selenium for 48 hours. The results found a 48-hour LC50 value of 839  $\mu$ g/l for selenium. There was 100% survival recorded in less than 5  $\mu$ g/l exposure of selenium, 96% survival recorded in 456  $\mu$ g/l exposure and 84% survival recorded in 985  $\mu$ g/l exposure (based on Table 2; Lussier *et al.*, 1998). Lussier *et al.* (1998) noted that selenium concentrations decreased by 11 to 14% during the experiment.

Marsden (2002) examined the effects of copper (Cu) spiked sediment on the survival and reproduction of *Paracorophium excavatum* collected from the Avon-Healthcote Estuary, Christchurch, through laboratory mesocosm experiments. Paracorophium excavatum was exposed to sediment spiked with 5 (control), 14, 20, 35 and 46 µg/g measured concentrations of Cu in experimental mesocosms for 28 days. The mesocosms contained seawater that was changed weekly. Four replicate copper exposures, and twenty Paracorophium excavatum individuals per replicate were used. At the end of exposure, the survival, growth, juvenile recruitment, and copper bioaccumulation were measured. The survival of Paracorophium excavatum decreased as copper concentrations increased. There was above 95% survival in 5  $\mu$ g/g and 14  $\mu$ g/g Cu exposures, above 75% survival in 20  $\mu$ g/g and 35  $\mu$ g/g Cu exposures. and 58% survival in 46 µg/g Cu exposure. The length of male Paracorophium excavatum significantly decreased with increasing sediment Cu concentrations, while there was no significant difference in female length. Juvenile recruitment was significantly reduced in sediments with 5, 14 and 20 µg/g of Cu, and no recruitment was observed in the highest Cu concentrations (35 or 46 µg/g). Reproduction was significantly affected by Cu-spiked sediment. No brooding females were observed in contaminated sediments after 28 days, but



in the control, female brood sizes were comparable with natural field populations. Cu
bioaccumulation in *Paracorophium excavatum* increased with increasing Cu concentrations.
Marsden (2002) concluded that low concentrations of Cu in sediment can affect the specific
life-history traits of *Paracorophium excavatum*, impairing growth, survival, and reproduction
enough to alter the population structure.

**McPherson & Chapman (2000)** investigated the effects of copper (Cu) on *Ampelisca abdita*, in laboratory-based static water-only toxicity tests. The study was conducted using field-collected amphipods which were acclimated in the laboratory. *Ampelisca abdita* was exposed to Cu concentrations ranging from 0.012 to 0.4 mg/l from a copper chloride stock solution, for 96 hours. There was one replicate for each treatment, and mortality was recorded daily. The 96-hour LC50 for *Ampelisca abdita* was 42.4 µg/l (based on nominal concentrations) and 33.5 µg/l (based on measured concentrations) of Cu. These results suggested *Ampelisca abdita* abdita had a higher sensitivity to Cu than *Eohaustorius estuarius*, another tested amphipod.

**Meadows & Erdem (1982)** examined the effects of mercury (Hg) on *Corophium volutator* found in sediment collected from Langbank, Firth of Clyde, Scotland. Marine animals and larger particles were removed from the sediment before it was exposed to 0.001 ppm, 0.1 ppm, 10 ppm and 1,000 ppm of mercury chloride in seawater. The sediment was divided into five parts; four parts of the sediment were washed six times with one of the Hg solutions and one part was washed with uncontaminated seawater to act as a control. After being washed in the Hg solution, the sediment was then placed into crystallising dishes which contained the same Hg-contaminated seawater as washed in, and between 30 to 40 *Corophium volutator* individuals. The seawater above the treated sediment was replaced on alternate days to make sure the Hg content did not vary appreciably. There were two replicate sets of six experiments conducted, each had different exposure durations lasting either 6 hours, 24 hours, 3 days, 7 days, 15 days, and 33 days. Mortality, LD50s and mercury uptake by the amphipod were recorded.

*Corophium volutator* mortality increased with Hg concentration and exposure time. All individuals died (100% mortality) within 6 hours of exposure to the highest concentration (1,000 ppm Hg). At 0.001 ppm Hg, a significant increase in mortality was seen between days 15 and 33. At 0.1 ppm and 10 ppm Hg, a significant increase was seen between days 7 and 15. Recorded LD50 values (Figure 1; Meadows & Erdem, 1982) decreased as Hg concentrations increased from around 30 days in 0.001 ppm Hg solutions and around 2.5 hours in 1,000 ppm Hg solutions. In the experiments, the sediment bacteria were also



counted as it is important for *Corophium valuator* habitat selection and an important food source. Declines in bacteria due to Hg exposure correlated with increases in *Corophium volutator* mortality. They reported that both living and dead amphipods accumulated large amounts of Hg, with accumulation higher in the dead amphipod individuals compared to the living. Meadows & Erdem (1982) concluded that the uptake of Hg and the progressive mortality of *Corophium volutator* will have taken place either by direct uptake of Hg across the body cuticle or by ingestion of contaminated bacteria and sediment during feeding.

Menchaca et al. (2010) investigated the effects of toxic sediment samples and reference toxicant cadmium (Cd) on laboratory-cultured and field-collected Corophium multisetosum. Corophium multisetosum from multiple life stages were collected from Bidasoa Estuary, and some of these individuals were maintained in controlled laboratory conditions for over a year. A liquid phase Cd toxicity test was carried out to specify seasonal sensitivity ranges. In this test, Corophium multisetosum were exposed to 0, 2, 5, 10 and 20 mg/l nominal concentrations of Cd for 72 hours. There were two replicates for each concentration and 15 Corophium multisetosum individuals per replicate. Mortality and measured concentrations of Cd were reported. The Cd tests were conducted twice, first in July 2008 with only fieldcollected amphipods and the second in August 2008 with field-collected and laboratorycultured amphipods. The measured Cd concentrations after 72 hours were 0.03, 1.53, 5.49, 11.37 and 21.06 mg/l Cd. Menchaca et al. (2010) found large variability in the sensitivity of the field amphipods according to the month they were sampled. The recorded 72-hour LC50 value in July (6.55 mg/l Cd) was higher than the 72-hour LC50 value recorded in August (2.40 mg/l Cd) for field Corophium multisetosum. The recorded 72-hour LC50 value for cultured Corophium multisetosum was 5.81 mg/l Cd. A significant difference was found between the LC50 values for the cultured and field amphipods in August.

In the sediment toxicity bioassays both field-collected and laboratory-cultured *Corophium multisetosum* were exposed to control sediment from Bidasoa Estuary and three different sediment samples categorized along a toxicity gradient; absence of toxicity (non-toxic), moderate toxicity and high toxicity. The sediment toxicity was related to high concentrations of copper (Cu) and zinc (Zn). Non-toxic sediment contained 81 mg/kg of Cu and 216 mg/kg of Zn, moderately toxic sediment, had 122 mg/kg of Cu and 414 mg/kg of Zn, and highly toxic sediment had 715 mg/kg of Cu and 1251 mg/kg of Zn. There were three replicates used for each sediment sample and the mortality and burial tendency were recorded after 10 days of exposure. The sediment tests were conducted twice, first in June 2008 with only field-collected amphipods and the second in August 2008 with field-collected and laboratory-



cultured amphipods. In control sediment there was no significant difference between mortality in cultured and field amphipods and there was above 90% survival. As predicted, the nontoxic sediment was not toxic to Corophium multisetosum in any sediment test. Field Corophium multisetosum from June and August exposed to the highly toxic and moderately toxic sediment had high mortalities, which were significantly different from the control sediment. There was 50% mortality recorded in the moderately toxic sediment and 83% mortality recorded in the highly toxic sediment (taken from Table 1). In contrast these results, the cultured Corophium multisetosum individuals did not respond to the toxic sediment and mortality (15%) was not significantly different from the control sediment. The cultured amphipods had lower mortality and therefore lower sensitivity to toxicity than field amphipods. A sublethal effect was also observed as the burial behaviour of the tested amphipods was assessed. There was a high burial tendency for both cultured and field-collected amphipods in the non-toxic and moderately toxic sediments. In contrast, cultured amphipods had a significantly lower burial rate in the highly toxic sediment. Menchaca et al. (2010) suggested several hypotheses, which could explain the differences in sensitivities in cultured and field Corophium multisetosum. It was suggested that the conditions under which cultured amphipods were maintained and the selection of individuals for tests could have influenced the responses observed in the experiments. In addition, the sensitivity of field amphipods could be related to multiple environmental stressors impacting their overall performance.

**Narracci et al. (2009)** conducted a cadmium (Cd) reference toxicant test, during a contaminated sediment toxicity test. In laboratory conditions, *Corophium insidiosum* was exposed to control seawater and 0.8, 1.6, 3.2 and 6.4 mg/l of cadmium chloride dissolved in seawater in aerated beakers for 96 hours. There were three replicates for each concentration treatment, and twenty individuals were placed in each beaker. The tests were conducted in spring-summer and autumn-winter, to test seasonal variations in *Corophium insidiosum* sensitivity to Cd. Narracci *et al.* (2009) found a mean 96-hour LC50 value of 1 mg/l of Cd in the summer-spring and 0.92 mg/l of Cd in the autumn-winter. There was no significant difference between seasons.

**Niera** *et al.* **(2014)** examined the effects of copper (Cu) contamination from boat moorings on benthic macrofaunal communities in three San Diego marinas, Southern California, USA. San Diego Bay has elevated Cu levels in the sediment and water column, which is associated with recreational boats, as more than 90% of boats in the bay have Cu-based antifoulant paint. Field sampling was conducted at 35 sites in three marinas: 15 sites in America's Cup (AC), 12 sites in Harbor Island West (HW) and eight Harbor Island East (HE)



to analyse the Cu concentrations in water (from bottom and surface) and sediments (porewater and solids). Sites were either described as 'with boats' (areas where boats are docked and densely concentrated) and 'without boats' (areas adjacent or opposite areas where boats dock. Cu sediment concentrations ranged from 120 mg/kg in samples from HE, 157.9 mg/kg in samples from HW and 215.6 mg/kg in samples from AC.

The Cu distribution in sediment followed a clear gradient, with the highest Cu concentrations in 'hotspots' directly beneath moored boats. Sediment cores were collected from 15 stations in AC, 12 stations in HW and eight stations in HE, and brought to the laboratory, where macrofauna were extracted from the sediment and analysed for macrofaunal abundance and biodiversity. Niera *et al.* (2014) reported that increased Cu sediment levels were associated with a decrease in species richness and shifts in community composition. Sites without boats had a greater abundance of *Corophium* sp. and other amphipods. Niera *et al.* (2014) noted that amphipods, including *Corophium* sp. appeared to be Cu-sensitive indicators.

**Pérez-Landa** *et al.* (2008) investigated the effects of cadmium (Cd) and total ammonia on the survival of *Corophium urdaibaiense* collected from the Urdaibai Estuary, Spain and *Corophium multisetosum* collected from the Bidasosa estuary, using 72-hour laboratorybased water only tests. The study conducted two sets of experiments to assess the effects of seasonality and body size on the sensitivity of the amphipods to both ammonia and Cd.

In the *Corophium urdaibaiense* seasonal ammonia experiments, the amphipod was exposed to 0, 36, 72, 110, 180 and 360 mg/l of ammonia in November 2002; March, May, July, August, and December 2003; and February 2004. Pérez-Landa *et al.* (2008) reported that *Corophium urdaibaiense* was less sensitive to ammonia in December (described as Autumn), with a 72-hour LC50 value of 110 mg/l ammonia. Sensitivity to ammonia increased in August, with a 72-hour LC50 value of 61 mg/l ammonia. *Corophium urdaibaiense* was more sensitive to ammonia in May (spring), with a 72-hour LC50 value of 27.1 mg/l ammonia.

In the *Corophium urdaibaiense* seasonal Cd experiments, the amphipod was exposed to 0, 1, 2, 4, 8, 12 mg/l of Cd in February and September 2002. Pérez-Landa *et al.* (2008) reported a 72-hour LC50 value of 2.29 mg/l of Cd in winter (February) and 0.88 mg/l of Cd in summer (September). Results from both *Corophium urdaibaiense* seasonal experiments show seasonal variability in LC50s. *Corophium urdaibaiense* was more sensitive to ammonia and Cd in the spring/summer and less sensitive in winter.



In the *Corophium multisetosum* seasonal ammonia experiments, the amphipod was exposed to 0, 36, 72, 110, 180 and 360 mg/l of ammonia in April, June, July, September and October 2004, and January 2005. Pérez-Landa *et al.* (2008) reported that *Corophium multisetosum* was more sensitive to ammonia in September (described as summer), with a 72-hour LC50 value of 25.6 mg/l ammonia and less sensitive in January (winter), with a 72-hour LC50 value of 115 mg/l ammonia.

In the *Corophium multisetosum* seasonal Cd experiments *Corophium multisetosum* was exposed to 0, 1, 2, 4, 8, 12, 16, 24, and 32 mg/l of Cd in April, June, July, September and October 2004, and January 2005. Pérez-Landa *et al.* (2008) reported that *Corophium multisetosum* was more sensitive to Cd in September (described as summer), with a 72-hour LC50 value of 0.63 mg/l Cd and less sensitive in January (winter), with a 72-hour LC50 value of 31.5 mg/l Cd. The results from both *Corophium multisetosum* seasonal experiments show seasonal variability in LC50s, as the amphipod was more sensitive to ammonia and Cd in summer and less sensitive in the winter.

In the body size experiments, *Corophium urdaibaiense* from three different size groups: large (2 to 4 mm), medium (1 to 2 mm) and short (0.5 to 1 mm) were exposed to 0, 36, 72, 110, 180 and 360 mg/l of ammonia in water for 72 hours. The small-sized group consisted of only juvenile amphipods, the medium-sized group consisted of juveniles and adults, and the large-sized group consisted of only adults. Pérez-Landa *et al.* (2008) reported a positive correlation between body size and *Corophium urdaibaiense* body size. The recorded 72-hour LC50 values were approximately 70 mg/l ammonia for the short-size group, approximately 80 mg/l ammonia for the medium-size group and approximately 90 mg/l ammonia for the large-size group (Taken from Figure 2, Pérez-Landa *et al.*, 2008). Overall, differences in sensitivities of *Corophium urdaibaiense* to ammonia were found between body size groups (Pérez-Landa *et al.*, 2008).

**Picone et al. (2008)** examined the sensitivity of *Corophium orientale* collected from Magra River estuary, Italy, to pure chemicals Nickel (Ni), Sodium Dodecyl-Sulphate (SDS) and reference toxicant cadmium (Cd) using laboratory-based water-only toxicity tests and to contaminated sediment from Venice Lagoon, using laboratory-based static sediment toxicity test. The amphipods were tested during a period from August 2003 to March 2004. In the water-only toxicity tests, juvenile and young adult *Corophium orientale* were exposed to five nominal concentrations of each toxicant and negative control in glass beakers for 96 hours; there were three replicates and 20 individuals per replicate. The reference toxicant test



exposed *Corophium orientale* to 0.8, 1.6, 3.2, 6.4 and 12.8 mg/l of Cd and recorded a mean 96-hour LC50 value was 3.3 mg/l Cd. The Cd sensitivity varied during the testing period, with the highest LC50 value (10.5 mg/l Cd) recorded in March 2004 (early spring) and the lowest LC50 value (1.44 mg/l Cd) recorded in September 2003 (late summer). The reason for seasonal variation is unknown.

The Ni test exposed *Corophium orientale* to 80, 160, 240, 320 and 400 mg/l of Ni. Picone *et al.* (2008) recorded a 96-hour LC50 value of 352 mg/l, LOEC value of 160 mg/l and NOEC value of 80 mg/l. The SDS test exposed *Corophium orientale* to 1, 2, 4, 8 and 16 mg/l of SDS. They recorded a 96-hour LC50 value of 8.7 mg/l, LOEC value of 4.0 mg/l and NOEC value of 2.0 mg/l. Overall, the results from the water-only tests showed Cd was the most toxic contaminant to *Corophium orientale*, followed by SDS then Ni.

In the contaminated sediment test, Corophium orientale was exposed to 50 sediment samples from the Lagoon of Venice and negative controls in glass beakers containing overlying artificial seawater for 10 days. There were three replicates and 25 individuals per replicate. Chemical analysis conducted in this study revealed the sediment is contaminated with a mixture of eight heavy metals (As, Cd, Cr, Cu, Hg, Ni, Pb and Zn) and has significant total organic carbon content. Previous literature noted the sediment sampling sites were also characterized by organic micropollutant concentrations. Picone et al. (2008) reported a range of mortalities from 12% to 62% in sites classified as toxic in the study developed site-specific toxicity scoring system. The worst-case mortality (62%) was reported in the only site described as having 'Extreme' toxicity. Picone et al. (2008) stated that the results displayed acute toxicity for the amphipod in only a few sites, which were located close to an industrial area and in deep sediments, indicating most of the sediment samples from the lagoon were not toxic toward the amphipod species. The chemical analysis of the sediment also showed no difference in sediment contamination, and it is unclear which contaminant contributed to toxicity. This study has shown a relatively low sensitivity for Corophium orinetale compared to other amphipod species such as Ampelisca abdita and Rhepoxynius abronius that are commonly used in toxicity tests

**Picone et al. (2016)** examined the effect of contaminated sediment from the Venice Iagoon, Italy, on *Corophium volutator*, using laboratory-based whole sediment mortality tests. Sediment samples were collected from 10 sampling sites within the Iagoon along a chemical contamination gradient, during three different seasonal campaigns: in May 2004 (spring campaign), October 2004 (autumn campaign) and February 2005 (winter campaign).



Chemical analysis revealed sediments were contaminated with a mixture of ammonia, sulphides, heavy metals, and PAHs. Adult *Corophium volutator* were exposed to the contaminated sediment samples and artificial seawater in test chambers for 10 days; there were five replicates and 25 individuals per replicate. No acute toxicity towards *Corophium volutator was* observed. Picone *et al.* (2016) concluded that the sediment contamination in the studied Lagoon site, especially regarding hydrocarbons and PAHs, was too low to induce mortality of *Corophium volutator*. In addition, a water-only positive cadmium (Cd) control test was carried out, which exposed *Corophium* volutator to 0.8, 1.6, 3.2, 6.4 and 12.8 mg/l of Cd for 10 days. The recorded 10-day LC50 values ranged from 1.0 to 6.7 mg/l of Cd. There was no significant seasonal variation between *Corophium volutator* sensitivity, with the highest LC50 value of 4.7 mg/l Cd found in October and the lowest LC50 value of 2.0 mg/l Cd found in February.

**Prato et al. (2006)** examined a number of sediment samples from the Mar Piccolo basin, Italy, using multiple bioassays with *Corophium insidiosum* and *Gammarus aequicauda*. Cadmium chloride was used as a reference toxicant in water-only assay. *Corophium* was exposed to 0.8, 1.6, 3.2 and 6.4 mg/l CdCl<sub>2</sub> for 96 hours. The resultant 96-hour LC50 varied seasonally from 0.35 to 3.36 mg/l CdCl<sub>2</sub>. *Gammarus* was exposed to 0.2, 0.4, 0.8, and 1.6 mg/l CdCl<sub>2</sub> for 96 hours. The resultant 96-hour LC50 varied seasonally was 0.26 to 5.16 mg/l CdCl<sub>2</sub>. LC50s were highest in between September and December in *Gammarus*. Mortality due to sediment exposure was significantly higher than controls at two stations, which were contaminated with metals (Cd, Cu, Hg, Ni, Pb, and Zn). Mortality at these stations were 34.6 and 51% in *Corophium* and 68.3 and 51.6% in *Gammarus*. Prato *et al.* (2006) noted that *Gammarus aequicauda* was more sensitive to the contaminated sediment than *Corophium insidiosum* but that metals may not have been the only contaminant present.

**Prato et al. (2008)** examined the effect of temperature and season on cadmium toxicity in *Corophium insidiosum. Corophium* were collected from the Mar Piccolo basin, Italy in August, November and January. Mortality in laboratory sediment was measured at 10, 15, 20 and 25°C for 10 days after 48-hour acclimation. They were exposed to CdCl<sub>2</sub> (0.8, 1.6, 3.2 and 6.4 mg/l) at the separate temperature above, for 96 hours in water-only assays. Mortality in control sediment ranged from 2.6% at 10°C in August to 17% at 20 in November. There were significant difference between the mortality in August and November samples at 25°C. The 96-hour LC50s decreased (Cd toxicity increased) as temperature increased and ranged from 2.11 mg/l Cd to 0.7 mg/l. The LC50s were highest at 10°C in November and January and at



15°C in August. Prato *et al.* (2008) concluded that 20°C was the optimum temperature for *Corophium* bioassays in summer and 15°C in other seasons.

**Prato et al. (2011)** examined the acute toxicity of the epiphytic dinoflagellate *Osteropsis* cfr. *ovata* to four crustacean species, including *Corophium insidiosum*. The study used water-only exposure to copper chloride as a positive control, to determine sensitivity when exposed to a single reference toxicant. They reported LC50 value of 1.06 mg/l of copper\_for *Corophium insidiosum*.

**Prato et al. (2012)** examined a number of sediment samples from the Mar Piccolo basin, Italy, using multiple bioassays based on several species, including *Corophium insidiosum*. Cadmium chloride was used as a reference toxicant in water-only assay. *Corophium* was exposed to0.4, 0.8, 1.6, 3.2 and 6.4 mg/l CdCl<sub>2</sub> for 96 hours. The resultant 96-hour LC50 was 1.15 (0.71 to 1.39) mg/l CdCl<sub>2</sub>. In whole sediment assays, the more contaminated inner inlet stations showed significantly higher toxicity (mortality) than the outer inlet stations. Mortality ranged from <5% to ca 35% across the samples examined (based on figure 2). However, the nature of the contaminants were not specified.

**Re et al. (2009)** reported the toxicity of the reference toxicant CdCl<sub>2</sub> (0.034, 0.061, 0.110, 0.196; 0.343, 0.613, 1.104, 1.962 mg Cd<sup>2+</sup>/l) to *Corophium multisetosum* in 96-hour assays in water-only. The resultant 96-hour LC50 was 0.33 mg Cd<sup>2+</sup>/l at 22°C and 0.57 mg Cd<sup>2+</sup>/l at 15°C. Re *et al.* (2009) also examined differences in the community and *Corophium* mortality due to exposure to samples of sediment from the Tagus estuary, Portugal before and after a significant flood event in 2000 to 2001. *Corophium* mortality decreased in sediments after the flood due to decreased organic contamination (DDT metabolites, PAHs, PCBs, HCH) and changes in sediment structure (e.g. grain size, redox) but no one causal factor was identified.

**Redmond** *et al.* (1994) investigated the sensitivity of field-collected *Ampelisca abdita* collected from the Pettaquamscutt River, Narragansett, Rhode Island and cultured *Ampelisca abdita* to cadmium chloride. In laboratory conditions, *Ampelisca abdita* was exposed to seawater spiked with 0 mg/l, 0.09 mg/l, 0.19 mg/l, 0.38 mg/l, 0.75 mg/l and 1.5 mg/l nominal concentrations of cadmium chloride for 96 hours. Cultured amphipods were collected from the same Pettaquamscutt River population but were collected on three different occasions. For each of these samples, three tests were conducted. Cultured amphipods had a 96-hour LC50 of 0.40 mg/l, 0.28 mg/l and 58 mg/l. The field collected amphipods had a 96-hour LC50 value of 0.20 mg/l. The authors concluded that the sensitivity of laboratory cultured



amphipods to cadmium was comparable to the field collected amphipods. They concluded that *Ampelisca abdita* was suitable for toxicity testing.

**Reish (1993)** examined the toxicity and bioaccumulation of metals (As, Cd, Cr, Cu, Hg, Pb, & Zn), DDT, a PCB (Arochlor 1254), and the water-soluble fraction (WAF) of diesel fuel in the amphipods *Corophium insidiosum* and *Elasmopus bampo*. Adult amphipods were exposed to a range of concentrations of each chemical for 96 hours in water only assays and mortality recorded. The resultant 96-hours LC50s in *Corophium* were 1.1 mg/l As, 0.68 mg/l Cd, 11.0 mg/l Cr, 0.6 mg/l Cu, >5.0 mg/l Pb, 0.02 mg/l Hg, 1.9 mg/l Zn, 0.00007 to 0.0004 mg/l DDT, 0.009 mg/l PCB, 0.9 mg/l WAF diesel. *Corophium insidiosum* was more sensitive to As, Zn, DDT, and PCB than *Elasmopus bampo*, while *Elasmopus bampo* was more sensitive to Cd, Cr, and Cu than *Corophium insidiosum*. Mercury, cadmium, and copper were the most toxic metals to amphipods. Both species of amphipod were very sensitive to DDT and the PCB, which agreed with other studies reviewed in this paper.

**Riba et al. (2003)** examined the toxicity of sediment, contaminated by heavy metals (As, Cd, Cu, Pb, Zn) after the Aznacollar mining spill (April 1998) in the Donana National Parks, Spain. *Ampelisca brevicornis* and *Corophium volutator* were exposed for 10 days to diluted contaminated sediment collected from the Guadalquivir estuary and mortality recorded. Contaminated sediment was diluted with control (clean) sediment to give dilutions of 0.3, 1.8 and 7.9%. Mortality at a dilution of 0.3% was 75 to 95% for *Ampelisca* and 60 to 80% for *Corophium*. Mortality increased to 90% for both species at a dilution of 1.8% and reached 100% for both species at a dilution of 7.9%. Riba *et al.* (2003) used the dilution factors to derive a 10-day LC50 of 0.67% dilution for *Ampelisca* and 0.56 % dilution for *Corophium* but it was unclear how this value could be related to the measured concentrations of the constituent metal contaminants.

**Roberts et al. (2013)** examined the toxicity of heavy metal (As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, Zn) contaminated vs. relatively uncontaminated reference sediment to *Corophium volutator* in the presence of pCO<sub>2</sub> to simulate ocean acidification. *Corophium* was exposed to containers of reference or contaminated sediment at 390, 750, 1140 µatm nominal pCO<sub>2</sub> for 9 days and mortality recorded. The mean survival of *Corophium* in the contaminated sediments at 390 and 750 µatm pCO<sub>2</sub> was 50 to 55% but was not significantly different from the survival in uncontaminated reference sediment. However, survival was significantly lower (6%) at 1140 750 µatm pCO<sub>2</sub>. Roberts *et al.* (2013) concluded that exposure to pCO<sub>2</sub> (ocean acidification)



increased the toxicity of the contaminated sediment, probably due to the effects of pH on their physiology rather than metal speciation.

**Shipp & Grant (2006)** examined the effectiveness of feeding in *Hydrobia ulvae* as a bioassay for contaminated sediment, in comparison to a standard bioassay using *Corophium volutator*. Both species were exposed to heavy metal contaminated sediment from the Fal and Hayle estuaries, Cornwall, using standard 10-days bioassay protocols (unspecified). The mortality (ca 30% based on Figure 3; Shipp & Grant, 2006) of *Corophium volutator* was significantly higher than controls when exposed to Restronguet Creek sediment where the sediment copper (Cu) concentration exceeded 2,000  $\mu$ g/g. But mortality was significant (ca 50% based on Figure 3) when exposed to Hayle estuary sediment where pore water Cu was only 260  $\mu$ g/g. Shipp & Grant (2006) suggested that *Corophium* was more exposed to Cu in pore water than in sediment because of it lives in U-shaped burrows.

**Strode & Balode (2013)** examined the effect of heavy metal exposure on Baltic amphipods, including *Bathyporeia pilosa*, in Latvian territorial waters of the open Baltic Sea and the Gulf of Riga using laboratory-based acute toxicity tests. Juvenile *Bathyporeia pilosa* were exposed to cadmium (CdCl<sub>2</sub>), copper (CuSO<sub>4</sub>), and zinc (ZnSO<sub>4</sub>·7H<sub>2</sub>O) in controlled conditions for 48-hour and 96-hour periods. The determined 48-hour LC50 values for *Bathyporeia pilosa* were 1.46 mg/l for cadmium, 1.74 mg/l for copper, and 3.53 mg/l for zinc and the 96-hour LC50s were 0.35 mg/l for cadmium, 1.07 mg/l for copper, and 1.15 mg/l for zinc. The study found that *Bathyporeia pilosa* exhibited moderate resistance compared to other tested amphipods, with the freshwater species *Gammarus pulex* being among the most sensitive. Strode & Balode (2013) concluded that sensitivity varied significantly between species (p<0.05), with cadmium being the most toxic that was tested. The findings highlight the need for species-specific ecotoxicological assessments in brackish environment and using native species for more accurate results, aligning with region specific environmental conditions.

**Tay et al. (1992)** assayed the effects of contaminated sediment from Halifax Harbour (Nova Scotia) on a selection of invertebrates, including the amphipods *Corophium volutator* and *Rhepoxynius abronius*. The sediment samples were contaminated by metals (As, Cd, Cu, Hg, Pb, and Zn) and 16 PAHs (Tables 2 & 3). Tay *et al.* (1992) reported 96-hour LC50s of 10.1 and 22.7 mg/l Cd in *Corophium volutator* and 0.9 and 1.9 mg/l Cd in *Rhepoxynius abronius*. The toxicity of the sediments varied between sites. Three sites were not toxic to *Corophium* but toxic to *Rhepoxynius*, while sediments from Sydney Tar Pond were acutely toxic resulting in 100% mortality in both species. The percentage mortality of *Rhepoxynius* 



was highest in sediments contaminated with high levels of PAHs (25.43 mg/kg), while *Corophium* was less sensitive and 91% survived exposure to sediment contaminated with 25.43 mg/kg PAH, although only 47.3% were burrowed at the end of the test. Tay *et al.* (1992) suggested that tube-dwelling *Corophium* were less exposed to the contaminants than the surface burrowing *Rhepoxynius*.

**Warwick (2001)** compared the invertebrate macrofauna of the Fal estuary to six other estuaries in the UK. Macrofaunal survey data was compared using multi-dimensional scaling. The Fal estuary is heavily contaminated by metals due to mining activity. The invertebrate macrofaunal communities of the Fal were significantly different from those of the other estuaries, primarily due to the complete absence of the amphipod *Corophium volutator* and the isopod *Cyathura carinata* and the abundance of opportunistic annelids. Warwick (2001) concluded that metal contamination was the likely cause of their absence.

## 4.2 Organometals

Only one article examined the effects of tributyltin (TBT) on Corophium.

**Stronkhorst et al. (1999)** studied the acute toxicity of tributyltin (TBT) on sediment dweller *Corophium volutator* collected from the Eastern Scheldt estuary, Netherlands, in laboratorybased TBT spiked sediment tests. Marine sediment was spiked with tributyltin chloride to gain nominal TBT concentrations of 0, 32, 100, 320, 1,000, 3,200, 10,000 ng/g dry weight sediment. The concentrations of TBT, dibutyltin (DBT) and monobutyltin (MBT) in spiked sediment and pore water were recorded. Pore water concentrations ranged from <3 to 1,636 ng/l. Twenty *Corophium volutator* were added to each jar containing spiked sediment and seawater, from the Eastern Scheldt estuary, for 10 days. The survival of *Corophium volutator* decreased between TBT sediment concentrations of 1,144 and 2,383 ng/g dry weight and pore water concentrations of 106 and 540 ng/l. The NOEC and 10-day LC50 values of TBT in sediment were 1,144 ng/g and 2,185 ng/g, respectively. The NOEC and 10-day LC50 values of TBT in pore water were 107 ng/l and 329 ng/l, respectively. The evidence from this study suggested that the survival of *Corophium volutator* was adversely affected at high sediment concentrations of TBT. The LC50 values indicated that TBT was very toxic to *Corophium volutator*.



## 4.3 Nanoparticulate metals

Only four articles examined nanoparticulate (NP) metals. All four examined the toxicity of zinc oxide nanoparticulates (ZnO-NP) and only one examined the toxicity of ZnO-NP and calcium phosphate nanoparticles (CaP-N). The evidence is summarized below.

**Fabrega et al. (2012)** investigated the effect of chronic waterborne exposure of zinc oxide nanoparticles (ZnO NP), bulk zinc oxide (ZnO) and soluble zinc (zinc chloride) on the lifecycle of *Corophium volutator* collected from Otter estuary, Devon. In laboratory conditions, 20 organisms were placed in glass beakers containing sediment and water, and exposed to 0.2, 0.5 and 1 mg/l of each form of zinc via dosing of the water for 100 days. There were nine beakers for each concentration treatment, three replicates and redosing occurred every seven days. Survival, growth rate (rate of increase in body length) and reproduction (number of neonates) were recorded at days 28, 63 and 100. All forms of zinc affected the survival of *Corophium volutator*, with greater effects at the highest exposure concentrations (1 mg/l) after 100 days. After 100 days, the percentage survival of *Corophium volutator* was significantly different from the percentage survival in the control. For example, after 100 days at 1 mg/l of ZnO NP there was 66.7% survival (33.3% mortality) and at 1 mg/l of ZnO and zinc chloride there was 63.3% survival (36.7% mortality).

Growth was also significantly affected by all forms of zinc, with the highest concentrations leading to reduced growth rates, delayed sexual maturity and reduced fecundity. At 23 days, the highest concentrations of ZnO NPs, bulk ZnO and zinc chloride reduced the growth rate by around 11%, 12% and 21%, respectively. By 63 days, slower growth rates were only observed in *Corophium volutator* populations exposed to ZnO and zinc chloride. After 100 days, most individuals had reached adult sizes, except ones exposed to 1 mg/l of bulk ZnO, which were still significantly smaller. It was suggested that exposure to sublethal concentrations of zinc could potentially impact populations by reducing reproductive fitness and overall reproductive output. Fabrega *et al.* (2012) concluded that ZnO NPs were no more toxic than the other forms of zinc. They emphasised that zinc bioavailability and toxicity were influenced by aggregation, dissolution, and sediment interactions, but caused comparable long-term effects.

**Larner** *et al.* **(2012)** examined the effects of three different forms of zinc oxide (ZnO): zinc oxide in nanoparticulate form (ZnO-NPs), bulk ZnO and soluble zinc (ZnCl<sub>2</sub>) on *Corophium volutator* collected from the Otter estuary, Devon UK. The ZnO-NPs were labelled with a single stable Zn isotope to trace Zn movement. In laboratory conditions, adult *Corophium* 



*volutator* was exposed to ZnO-NPs (around 1,000 ng/g Zn), bulk ZnO (around 750 ng/g Zn) and soluble ZnCl<sub>2</sub> (250 ng/g Zn) in water for 10 days. The uptake of Zn by *Corophium volutator* was analysed. *Corophium volutator* accumulated Zn from the dissolution of ZnO particles. In addition, there was more than 80% survival recorded in *Corophium volutator* exposed to ZnO-NPs and bulk ZnO concentrations and 70% survival recorded in *Corophium volutator volutator* exposed to soluble ZnCl<sub>2</sub>. The evidence reported low mortality, suggesting the amphipod species was able to tolerate the different Zn concentration exposures tested in this study.

**Righi** *et al.* (2023) examined the toxicity of zinc oxide (ZnO NPs) and calcium phosphate nanoparticles (CaP-N) using bioassays with an alga, copepod, and two amphipods *Corophium insidiosum* and *Gammarus aequicauda*. They also examined the environmental impacts of the two nanoparticles via a life cycle assessment (LCA). Juvenile amphipods were exposed to a range of concentrations of each nanoparticle in 96-hour bioassays and the relevant LC50s determined. They reported a 96-hour LC50 of 1.93 mg/l ZnO-NP in *Corophium* and 0.32 mg/l ZnO-NP in *Gammarus*. All LC50s for CaP-N were >100 mg/l and considered non-toxic. Righi *et al.* (2023) also noted that *Corophium insidiosum* constructed their tubes using the CaP-N nanoparticles during the 96-hour experiment, which they suggested also indicated CaP-N were non-toxic.

**Vimercati et al. (2020)** examined the toxicity of ZnO NPs (zinc nanoparticles) and ZnSO<sub>4</sub> on immature adult *Corophium insidiosum* and larval (nauplii) *Tigriopus fulvus*. The specimens were exposed to a range of concentrations (0.4, 0.8, 1.6, 3.2, & 6.4 mg/l) of each chemical, for up to 96 hours, with three replicates, mortality recorded at 24-hour intervals, and 96-hour LC50s determined. The 96-hour LC 50 for ZnO NPs was 1.75 mg/l, and for ZnSO<sub>4</sub> was 1.63 mg/l. They were not statistically significant. The average relative mortality of ZnSO<sub>4</sub> was slightly but not significantly higher than that of ZnO NPs. The acute mortality after 96 hours at the highest concentration 6.4 mg/l of ZnSO<sub>4</sub> was 100% while the acute mortality of ZnO NPs was 97.8%. Vimercati *et al.* (2020) concluded that the toxicity was due to zinc ions and that dissolution of the ZnO NPs was a crucial contribution to their toxicity.



## 5 Synthetic compounds – including Pesticides and Pharmaceuticals

A total of 83 results (ranked 'worst-case' mortalities) were obtained from 35 articles that examined the effects of 'Synthetic compounds' on amphipod species. Pesticides/biocides were most studied, with 74.4% of the results, followed by 'Synthetics (other)' (8.5%) and PCBs (6.1%) (Figure 5.1).

## 5.1 Pesticides/biocides

Where possible the pesticides/biocides were categorised by their function or target, for example herbicides, insecticides, rodenticides, or acaricides. The majority of results (worst-case ranked mortalities) for the effects of pesticides/biocides were from studies of organohalogens (22.9%), herbicides (18%), carbamates (18%) and parasiticides (14.75%) (Figure 5.1). The evidence is summarized below.

Allen *et al.* (2007) examined the effects of the parasiticide Ivermectin (IVM) on *Corophium volutator* juveniles, through 10-day acute toxicity tests and 28-day sublethal sediment tests in laboratory conditions. Amphipods were collected from the River Crouch and acclimated in an aquarium supplied with running, filtered clean seawater and a layer of clean reference sediment. The base sediment used in all tests was collected from Shoebury Sands, Southend. In the acute toxicity tests, *Corophium volutator* was exposed to nominal test concentrations ranging from 5 to 105  $\mu$ g IVM/kg wet sediment for ten days. Approximately ten adults were added to each acute test treatment and mortality was recorded as an endpoint. The 10-day tests were conducted on two occasions, two years apart. The results found 10-day LC50 values of 22.0  $\mu$ g/kg IVM in the first-year test and 21.9  $\mu$ g/kg IVM in the second test. The test results were essentially the same despite being conducted two years apart.

In the sublethal test, juvenile *Corophium volutator* were exposed to nominal test concentrations ranging from 0.5 to 29  $\mu$ g IVM/kg wet sediment for 28 days. There were three replicates for each experiment. Mortality and growth were determined by measuring the increase in length from head to telson, and dry weight. The results found using juveniles and longer exposure time did not increase *Corophium volutator* sensitivity to IVM. The 28-day LC50 value was 16.7  $\mu$ g/kg IVM. This was not significantly different from the 10-day LC50 value.





Figure 5.1. Count of ranked worst-case mortalities due to exposure to 'Synthetic compounds' in selected amphipods. Mortality is ranked as follows: 'Severe' (>75%), 'Significant' (25-75%), 'Some' (<25%), 'None (reported)', and 'Sublethal' effects.

In the highest concentrations of IVM, mortality was high but surviving *Corophium volutator* had faster growth rate (in weight terms) than the controls. The growth rate in the control was lower than in the IVM treatments. Therefore, the 28-day LOEC for *Corophium volutator* 



growth was more than 29 µg IVM/kg wet sediment. The authors concluded that IVM exerts deleterious effects on non-target organisms under and around fish farms where the parasiticide is used, but it also has the potential to persist for a very long time in the sediment. The evidence presented shows that IVM is acutely toxic to *Corophium volutator* in very low sediment concentrations. No definitive conclusions could be made on the sublethal toxicity of IVM on *Corophium volutator* growth rates, and further research is necessary (Allen *et al.,* 2007).

Ciarelli et al. (1997) examined the effects of lindane and cadmium on Corophium volutator, through laboratory-based static sediment toxicity tests. Corophium volutator collected from a clean intertidal area in Oesterput, The Netherlands, was exposed to sediment spiked with nominal concentrations 0.32, 0.57, 1.03, 1.86 and 3.35 µg/g (dry weight) of lindane (dissolved into acetone) and a solvent control with un-spiked sediment for 10 days. Three replicates were used, and twenty amphipods were in each replicate. The percentage of mortality was recorded. The spiked sediment experiments were conducted at regular intervals on seven occasions from July 1993 to April 1995. Four experiments used random Corophium volutator individuals, and three experiments were carried out using Corophium volutator individuals of three different lengths (5, 7 and 8 mm) to investigate if differences in sensitivity of lindane changes with body size or seasonally. The recorded 10-day LC50 values ranged between 0.78 and 1.49 µg/g of lindane. The results found seasonal differences in Corophium volutator responses to lindane. The highest values were recorded in April 1994 (1.46 µg/and April 1995 (1.49  $\mu$ g/). The July 1994 test showed a significant difference from all other tests (n = 6) at the lowest lindane concentration (0.32  $\mu$ g/g) and was also significantly different from the January 1994, January 1995, and April 1995 tests at 0.57 µg/g. The November 1994 test differed significantly from only the April 1994 test at 1.03 µg/g and from both the April 1994 and April 1995 tests at 1.86 µg/g. At the highest concentration (3.35 µg/g), the April 1995 test significantly differed from all other tests. Their results indicated that sensitivity was lowest in April. There were no significant differences in the sensitivities between the three different size classes, indicating that sensitivity to lindane was not size-related. The 10-day LC50 values were 1.31  $\mu$ g/g for the 5 mm individuals, 1.28  $\mu$ g/g for the 7 mm individuals, and 1.26  $\mu$ g/g for the 8 mm individuals.

The study also conducted positive control tests using cadmium chloride as a reference toxicant to assess the quality and health of the tested *Corophium volutator* populations. *Corophium volutator* was exposed to 1, 1.8, 3.6 and 7.5 mg/l of cadmium chloride in the aqueous phase for three days. Mortality was recorded and the results were compared to



those obtained for lindane. There were four cadmium experiments conducted, one experiment with random *Corophium volutator* individuals and three experiments using *Corophium volutator* individuals of three different lengths (4, 6 and 7 mm). The 72-hour LC50 values ranged from 2.74 to 9.9 mg/l of total cadmium. Seasonal differences were also found in this cadmium study, as the highest 72-hour LC50 values were found in April 1994 (9.9 mg/l) and April 1995 (6.88 mg/l). Similarly to the lindane study, there were no significant differences found in the LC50 values of the three different size classes. The 72-hour LC50 values were 2.60 mg/l for the 4 mm individuals, 2.83 mg/l for the 6 mm individuals and 2.59 mg/l for the 7 mm individuals.

Ciarelli *et al.* (1997) concluded that the difference between the highest and lowest LC50 values over all the lindane-spiked sediment experiments did not exceed a factor of two. Ciarelli *et al.* (1997) stated that despite differences in percentage mortality data found between tests, the overall variability in the sensitivity of *Corophium volutator* to lindane was small and within a narrow range.

Collier & Pinn (1998) investigated the toxic effects of ivermectin on Corophium volutator, through laboratory-based sediment experiments. Intertidal sediment cores, naturally containing Corophium volutator and other benthic fauna, were collected from the Ythan Estuary, Aberdeen. Concentrations of 1, 10 and 100 µl/g aliquots of ivermectin were mixed with mud and seawater before being applied to the sediment surface, yielding final ivermectin concentrations of 0.8, 8.0 and 80.0 mg/m<sup>2</sup>. The experiment was conducted in eight tanks, with two replicates for each ivermectin concentration and two control tanks with no ivermectin added. The cores were sampled at regular intervals over 21 days and the benthic community assessed. For Corophium volutator, the abundance, biomass and body lengths were recorded. Collier & Pinn (1998) reported that the benthic community present in the sediment cores were dominated by Corophium volutator along with, Hediste diversicolor, Hydrobia ulvae and Oligochaetae. There was a decline in biomass and abundance of Corophium volutator with time in the ivermectin-contaminated sediment and the controls. The rate of decline increased with increasing ivermectin concentrations. Statistical tests found a significant difference in Corophium volutator abundance with time and ivermectin concentration but no significant interaction between concentration and time. Corophium volutator mortality was greatest during the first seven days of the experiment. The level of mortality increased as the level of contamination increased, with 100% mortality occurring in 14 days at concentrations of 80.0 mg/m<sup>2</sup> of ivermectin. However, it was noted that there was moderately high mortality in the control, suggesting the Corophium volutator community was



under stress in the laboratory conditions without ivermectin. In addition, ivermectin exposure altered the *Corophium volutator* population structure over time and at lower concentrations of ivermectin. Smaller individuals were more sensitive than larger-sized individuals. After two days, individuals of smaller size classes (3-4 mm and 7-8 mm body length) declined in abundance relatively to the controls, particularly at the highest ivermectin concentrations (8.0 and 80.0 mg/m2) but other size classes increased in growth. After 14 and 21 days, a decrease in abundance was observed for all individual sizes at all concentrations of ivermectin. This was greatest at the highest concentrations of ivermectin. Collier & Pinn (1998) concluded that both the level of ivermectin contamination and duration of exposure have a significant effect on the benthic community, and these changes occurred more rapidly at higher concentrations of ivermectin. *Corophium volutator* was found to be sensitive to ivermectin, but *Hediste diversicolor* was the most sensitive species tested.

**Davies et al. (1998)** investigated the effects of ivermectin on *Corophium volutator*, through laboratory-based whole sediment acute toxicity tests. In the study, sediment from Munlochy Bay, Inverness was exposed to 0.15, 0.35, 0.7, 1.5, 3.5 and 7.0 mg ivermectin /kg dry sediment in test beakers for 10 days in an initial definitive test. Twenty individual *Corophium volutator* were randomly introduced to each beaker; there were three replicate beakers per concentration and five replicate controls. They determined a 10-day LC50 value of 0.18 mg/kg. The maximum ivermectin concentration that caused no mortalities above the control was estimated to be 0.7 mg/kg at 24 hours and 0.15 mg/kg at 48 hours. An additional test was carried out to determine the NOEC value *Corophium volutator* was exposed to 0.0125, 0.025, 0.05, 0.1, 0.2 and 0.4 mg ivermectin /kg dry sediment in test beakers for 10 days. There were four replicate beakers per concentration and six controls. They found that 0.1 mg/kg of ivermectin was the lowest concentration to show a statistically significant difference in mortalities from the control. Therefore, the recorded LOEC was 0.1 mg/kg of ivermectin. The recorded NOEC was 0.05 mg/kg, which was the highest concentration to have no observed effect that was significantly different from the controls.

In addition, Davies *et al.* (1998) investigated the degradation of ivermectin and monitored its toxicity to *Corophium volutator*. In the degradation study, sediment was spiked with nominal concentrations of 0.1, 0.2, 0.4, 0.8 and 1.6 mg/kg of ivermectin in beakers placed in tanks supplied with clean seawater for 100 days. On days 0, 25, 50, 75 and 100, three beakers were removed and used for a 10-day whole sediment experiment with *Corophium volutator*. The results found no significant decrease in toxicity of ivermectin-treated sediment for up to 75 days. However, at day 100, toxicity had decreased by about 50%. This suggested that



ivermectin degraded in the sediment over time, and ivermectin has an estimated half-life of over 100 days. The full list of cumulative percentage mortalities for each ivermectin concentration from the definitive test and NOEC test and the recorded LC50 values from the degradation study can be seen in the evidence summary spreadsheet.

Dumbauld et al. (2001) investigated the impact of the pesticide carbaryl on benthic communities. The pesticide was used to manage the invasive ghost shrimp Neotrypaea californiensis and mud shrimp Upogebia pugettensis that invade Pacific oyster (Magallana gigas) farms in Willapa Bay, Washington. The experiments were carried out in the field at locations in Willapa Bay where the invasive shrimp are found in high densities. An initial small-scale experiment used four sets of replicate 100 m<sup>2</sup> treatments and control plots at two sites in the Palix River sub estuary and the Cedar River sub-estuary. A small hand sprayer was used to apply various concentrations of carbaryl (0, 0.4, 1.5 and 5.6 kg/ha) to the plots, but samples of benthic community abundance were only taken in the control (0 kg/ha) and 5.6 kg/ha treatments. The benthic fauna was sampled using sediment cores taken from each plot 24 hours, two weeks and one month after the spray. The community composition differed between sites, with higher species diversity in the Cedar River site, while the Palix River had fewer taxa. In general, carbaryl treatment only caused significant diversity changes at the Palix River site one month and one year after the spray. Crustaceans were the second most numerically important group of organisms found at the Cedar River (representing 22% of individuals), and the third most important group at the Palix River (representing 10% of individuals). Crustaceans also found to have the most significant negative response to carbaryl treatment. The amphipod Corophium acherusicum was found in the Cedar River sample site, with an average of 1,100 individuals collected in the untreated control plots but no Corophium acherusicum were found in the untreated control Palix River sample sites. There was no significant effect of carbaryl treatment on Corophium acherusicum except one year after the spray when its density was enhanced on the treated plots.

A large-scale experiment was conducted using two pesticide treatment sites in the Palix River sub-estuary, exposed to 8.4 kg/ha of carbaryl applied via helicopter and one control site over 300 m away from the nearest treated site. Nine samples were collected along a transect at each site, two days before carbaryl exposure, two days post-treatment, 51 days posttreatment. An additional set of four samples was taken one- and two-years post-treatment. The results of the large-scale experiment were similar to the results of the small-scale experiment, with the numerically dominant taxa of arthropods and molluscs being the same as found on the small-scale Cedar River plots. Crustaceans were more abundant than
molluscs in all three sites, representing 79% to 95% of organisms. *Corophium acherusicum* was one of the most numerically dominant organisms counted and was the only organism to be significantly affected by carbaryl application in comparison with pre-spray and two days post-spray samples. The abundance of *Corophium acherusicum* decreased by two orders of magnitude post-spray but recovered dramatically after 51 days post-treatment. After one- and two-years post-treatment, *Corophium acherusicum* declined at the control site, but declines were less significant in treated sites.

Dumbauld *et al.* (2001) concluded that *Corophium acherusicum* was affected by treatment of carbaryl but differences between treated and untreated sites were not significantly different. The authors noted that the lack of a statistically significant effect was not conclusive, since the tests statistical power due to low sample size and high variability observed in the data, but it does indicate that acute toxicity was not absolute. In the large-scale experiment, the effect of carbaryl was highly significant, with a 97% decrease in abundance two days postspray. The results of this study also indicate that *Corophium acherusicum* was able to recover to pre-spray abundances or enhanced abundances in carbaryl treated sites after one or two years.

**Felmer** *et al.* (1995) examined the effects of pesticides fenvalerate and endosulfan and the size of microcosm on estuarine benthic macroinvertebrate community structure and recolonization. The study conducted two (one for fenvalerate and one for endosulfan) sixweek laboratory experiments, using 32 microcosms, 16 each of small and large sizes. Sediment collected from Perdido Bay, Florida was spiked with 5.0  $\mu$ g/g and 50.0  $\mu$ g/g nominal concentration of fenvalerate or 1.0  $\mu$ g/g and 10.0  $\mu$ g/g nominal concentration of endosulfan and placed into the microcosms. There were six control microcosms, five low pesticide concentration microcosms and five high pesticide concentration microcosms. Seawater from Santa Rosa Sound, containing larval and juvenile invertebrates, was continuously pumped into the tanks to allow organisms to recolonize the sediment. Changes in pesticide levels in the sediment were tracked over time. The total number of organisms, total number of taxa and dominant taxa were recorded as indicators of effects. The amphipod, *Corophium acherusicum*, dominated in abundance in both experiments.

In the fervalerate experiment, *Corophium acherusicum* was highly abundant in the control and the 5.0  $\mu$ g/g fervalerate treatments, with an average abundance of 188.8 in the controls and 128.4 in the low treatments. This was not a significant difference. However, a significant difference was found between the average abundance in low and higher fervalerate



concentrations, with an average abundance of 15.4 in 50.0  $\mu$ g/g treatments. The average abundance of *Corophium acherusicum* significantly decreased with increasing fenvalerate concentrations. In the control, 5.0  $\mu$ g/g, and 50.0  $\mu$ g/g (small microcosm only) fenvalerate treatments, *Corophium acherusicum* was the most dominant species recorded. However, in the larger microcosm treatments with the highest fenvalerate concentrations *Corophium acherusicum* was the second most dominant. Felmer *et al.* (1995) noted that *Corophium acherusicum* acherusicum accounted for most of the decrease in abundance in the benthic community in response to fenvalerate.

Similar results were observed in the endosulfan experiment, which found that *Corophium acherusicum* dominated abundance in most treatments, apart from the highest endosulfan concentration (10.0  $\mu$ g/g) where it was replaced by *Molgula manhattensis*. *Corophium acherusicum* abundance decreased with increasing endosulfan concentrations, with an average abundance of 65.5 in controls, 43.4 in 1.0  $\mu$ g/g endosulfan, and 26.0 in 10.0  $\mu$ g/g endosulfan in small microcosms and 38.5 in controls, 51.0 in 1.0  $\mu$ g/g endosulfan, and 18.2 in 10.0  $\mu$ g/g endosulfan in large microcosms. The changes in abundance were not significant. In addition, fenvalerate and endosulfan concentrations decreased in the sediments at all experimental concentrations. Felmer *et al.* (1995) concluded that microcosm size had some measurable effects on the benthic community structure in response to the pesticides, as the decrease in sensitive taxa, such as *Corophium acherusicum*, was similar across different microcosm sizes.

**Felmer et al. (1997)** examined the effects of pesticide chlorpyrifos and microcosm size on benthic macroinvertebrate colonization. The study conducted a 42-day laboratory-based experiment using 32 microcosms, 16 each of small and large sizes. Sediment collected from Perdido Bay, Florida was spiked with nominal concentrations of 1  $\mu$ g/g and 10  $\mu$ g/g (wet sediment) chlorpyrifos and placed into the microcosms. There were six control microcosms, five low pesticide concentration microcosms and five high pesticide concentration microcosms. Seawater from Santa Rosa Sound, containing larval and juvenile invertebrates, was continuously pumped into the tanks to allow organisms to recolonise the sediment. Changes in pesticide levels in the sediment, organism density and the dissimilarity amongst treatments were analysed. Overall, the density of organisms decreased with increasing chlorpyrifos concentration. The amphipod *Corophium acherusicum* dominated abundance in all treatments apart from the highest concentration of chlorpyrifos (10  $\mu$ g/g). The average density of *Corophium acherusicum* was 285.8 in the controls, 88.5 in the 1  $\mu$ g/g treatment and 43.9 in the 10  $\mu$ g/g treatment. There were significant effects between the densities in the



control and low chlorpyrifos concentrations, but not between the low and high chlorpyrifos concentrations. There were on average higher densities of *Corophium acherusicum* in smaller microcosms than in larger microcosms. Felmer *et al.* (1997) found that variations in *Corophium* acherusicum (and *Neauthes succinea*) density contributed the most to the average percent dissimilarity in the community structure in high chlorpyrifos concentrations. Dissimilarity showed how much the community structure changes under the different experimental conditions. Felmer *et al.* (1997) concluded that chlorpyrifos and microcosm size significantly affected the benthic community structure.

Ferraro & Cole (1997) examined the effects of DDT (Dichlorodiphenyltrichloroethane) and its metabolites on benthic macrofauna, specifically amphipods including Corophium heteroceratum and Ampelisca abdita, using a field collected sediment from San Francisco Bay, California, USA. Samples of macrofauna in sediment were collected from 11 stations, along a DDT contaminated gradient: four stations in the Lauritzen Channel (stations 1 to 4), two in the Santa Fe Channel (stations 5 and 6), three in the Richard Harbor Channel (stations 7 to 9) and two in open San Francisco Bay (stations 22 and 23). Sediment samples were collected from the field stations using van Veen grabs and cores. In laboratory conditions, concentrations of DDT and its metabolites in the sediment samples were measured, and normalised to organic carbon, which is believed to be a better reflection of the bioavailability of sediment-associated organic compounds. In addition, the macrofauna was separated from the sediment for identification and analysis. Stations 1 to 3 had the highest DDT sediment concentrations, followed by station 4, then station 6, then station 5 and 7, with station 9 having the lowest DDT sediment concentration. The measured DDT and its metabolite concentrations ranged from 1  $\mu$ g/g organic carbon in station 9 to 8,826  $\mu$ g/g organic carbon in station 1. The results found that Ampelisca abdita and Corophium heteroceratum were rare or absent at sites with moderate to high DDT sediment concentrations. The amphipods were sensitive to DDT-contaminated sediment, as the populations reduced in abundance or were entirely absent. Both amphipod species were more abundant at sites 9, 22 and 3, which had the lowest DDT concentrations (taken from Table 4). The statistical analysis confirmed a negative correlation between DDT and amphipod abundance, with a decline of around 0.4 log units in amphipod density per log unit increase in DDT concentration. Ferraro & Cole (1997) concluded that high concentrations of DDT and its metabolites contaminated in sediments were toxic to amphipods.

Fox et al. (2014) examined the effects of the antifoulant medetomidine on *Corophium volutator* collected from the Avon River, Devon, United Kingdom, through two laboratory-



based experiments, a 28-day survival and growth study and a 76-day growth and reproduction study. Juvenile Corophium volutator (less than seven days old) were exposed to sediment spiked with nominal medetomidine concentrations of 1.0 µg/kg, 3.2 µg/kg, 10 µg/kg, 32 µg/kg, and 100 µg/kg (dry weight). There were 11 replicates of each concentration, three for the survival and growth study and six for the reproduction study, with 30 juveniles per replicate. During the experiments, overlying water was renewed with fresh dilution water every seven days. In the 28-day study, the number of Corophium volutator individuals, their weight and individual lengths were measured. In the 76-day study, the number of Corophium volutator was measured, individuals less than 3 mm were recorded as neonates, and the number of gravid females was recorded. Survival of Corophium volutator in all exposure treatments ranged from 47% to 100% in the 28-day survival and growth study. Significant mortality was observed in the highest medetomidine concentrations compared to the controls, with between 60 to 70% survival recorded in 32 µg/kg treatments and between 50 to 60% survival in 100 µg/kg treatments (taken from Figure 1). The recorded 28-day NOEC value for survival was 10 µg/kg and the recorded 28-day LOEC value for survival was 32 µg/kg. There was no significant effect of medetomidine exposure on weight or body length of juvenile Corophium volutator.

The survival of *Corophium volutator* in exposure treatments ranged from 27% to 97% in the 76-day reproduction study. There was significant mortality observed in the highest medetomidine concentrations compared to the controls, with between 50 to 60% survival recorded in 32  $\mu$ g/kg treatments and between 30 to 40% survival in 100  $\mu$ g/kg treatments (taken from Figure 2). The recorded 76-day NOEC value for survival was 10  $\mu$ g/kg and the recorded 76-day LOEC value for survival was 32  $\mu$ g/kg. There was a significant decrease in body weight and length of *Corophium volutator* after 76 days in the highest medetomidine concentration (100  $\mu$ g/kg) compared to the controls. No significant effect was found on the number of neonates between medetomidine treatments, but there was a significant decrease in the number of gravid females in the 100  $\mu$ g/kg medetomidine concentrations compared with the control. Fox *et al.* (2014) concluded that long-term exposure to medetomidine in sediments affected growth and reproduction (as a reduction in the number of gravid females) in *Corophium volutator* but only at a concentration at which survival is already compromised.

**Hellou et al. (2009a)** studied the effects of the pesticide Chlorothalonil and freshwater exposure on the behaviour and survival of *Corophium volutator* from the Bay of Fundy, through laboratory based spiked sediment and spiked seawater experiments. In the first experiment, *Corophium volutator* was exposed to sediment spiked with 0, 0.001, 0.1, 0.1, 1,



10, 100 ng/g of chlorothalonil for 48 hours. Exposure tanks containing the sediment were partitioned down the middle, which had a contaminated side of the sediment and the control reference side, which had less than 1 ml of acetone solution containing chlorothalonil, which could evaporate. The partition was removed, and individuals were placed in the centre of the tank. At the end of the experiment, individuals at each side were counted. The survival between exposures varied between 80 to 100% with a mean of 82% and they reported an LC50 value at 1,250 ng/g chlorothalonil. The amphipods were predicted to distribute within a 30% to 70% range between the two sides of the tank. For the intermediate levels of chlorothalonil, 2.5, 12.5 and 125 ng/g, there was a 30 to 50% difference between the two sides, with a tendency for the amphipod to prefer the contaminated side if it started on that side. The overall behavioural response formed a U-shape, with a shift towards reference sediment preference at higher pesticide levels (more than 1,250 ng/g).

The second experiment exposed ten *Corophium volutator* to seawater spiked with 0, 0.001, 0.1, 0.1, 1, 10, 100 ng/ml of acetone solutions containing chlorothalonil for 48 hours of exposure. The number of swimming individuals was counted after 24 and 48 hours of exposure. In the control, a mean of 25% of *Corophium volutator* were observed to be swimming, which reduced to around 10% of the individuals observed swimming in the lowest chlorothalonil concentration (0.001 ng/ml). There was no swimming observed in the highest concentration of chlorothalonil (100 ng/ml). Survival varied between 65 % to 100% and was not correlated with exposure level. Hellou *et al.* (2009a) concluded that behavioural effects were apparent in *Corophium volutator* in chlorothalonil exposures at 0.001 to 0.01 ng/g and at 0.001 to 0.01 ng/ml. In higher chlorothalonil concentrations above 100 ng/g and 100 ng/ml escape responses and reduced swimming were observed consistently, and less so in the lower concentrations.

**Hellou et al. (2009b)** studied the effects of pesticides, atrazine (AT), azinphos-methyl (AZ), carbofuran (CA) and endosulfan (EN) in sediment on the survival and behaviour of *Corophium volutator* collected from the Bay of Funday, Hantsport and Avenport, Nova Scotia Canada, in laboratory conditions over two years. In the first year, exposures were set up in tanks with separate compartments on each side for pesticide-spiked sediment and reference control sediment, partitioned by glass to separate the two sides. Once the partition glass was removed 20 *Corophium volutator* was added to the spiked sediment side in one tank and the reference side in a second tank. This experiment was conducted for all four pesticides. The spiked sediment had pesticide concentrations ranging from 0.004 to 250,000 ng/g in sediment for all four pesticide experiments. The exposure duration was two days. In the



second year, the method was refined as *Corophium volutator* individuals were added to the water column in the tanks rather than placed on the sediment, this enabled identical duplicates. In the second year, spiked sediments had pesticide concentrations of 0.02, 0.10, 0.50, 2.5 and 12.5 ng/g. The exposure duration was two days. The 2-day LC50 values varied between the pesticides. Both CA and AZ had 2-day LC50 values of 73 ng/g and LC20 values of 41 ng/g, EN had a 2-day LC50 value of 232 ng/g and LC20 value of 130 ng/g. AT concentrations had the highest LC50 value of over 250,000 ng/g and LC20 value of 487 ng/g, indicating AT was the least toxic pesticide. The study observed that only EN exposure caused significant avoidance behaviour, as *Corophium volutator* actively moved away from the spiked sediment towards the uncontaminated reference sediment. Hellou *et al.* (2009b) concluded that pesticide partitioning significantly determines toxicity and behavioural responses in *Corophium volutator*.

**Hyne et al. (2002)** examined the acute toxicity of four selenium compounds on adult and juvenile *Corophium* sp., collected from Lack Macquarie, through laboratory-based water-only and sediment toxicity tests. The selenium compounds used in the study were highly soluble and oxidation forms, selenite, and selenate; and organic seleno-amino acids, seleno-L-methionine and seleno-DL-cystine. In the water-only experiments, juvenile *Corophium* sp. were exposed to a range of 0.1 and 100  $\mu$ g/l of all four selenium compounds dissolved in water for 96 hours. The test solutions were renewed throughout the duration of exposure. Each experiment was replicated two or three times, and each test beaker contained 10 juveniles.

Juveniles were more sensitive to seleno-L-methionine and seleno-DL-cystine than selenite and selenate, with recorded 96-hour LC50 values of 1.5  $\mu$ g/l for seleno-L-methionine and 12.7  $\mu$ g/l for seleno-DL-cystine. The recorded NOEC value was 0.25  $\mu$ g/l and the LOEC value was 0.87  $\mu$ g/l in seleno-L-methionine exposure. The recorded NOEC value was less than 1.2  $\mu$ g/l and the LOEC value was 1.2  $\mu$ g/l in seleno-DL-cystine exposure. On the other hand, selenate and selenite were less toxic to juvenile amphipods, with NOEC values of 116 and 58  $\mu$ g/l, respectively. Adult *Corophium* sp. was exposed to 0.1 to 9.3  $\mu$ g/l final measured concentrations of selo-L-methionine dissolved in seawater for 96 hours. The test solutions were renewed daily, and each experiment was replicated three times. Each test beaker contained 10 adults. Results were similar to those recorded in the juvenile water-only experiment, with a recorded 96-hour LC50 value of 2.1  $\mu$ g/l and NOEC value of 0.70  $\mu$ g/l.



In the sediment toxicity tests, juvenile and adult *Corophium* sp. were exposed to sediment spiked with 0, 0.3, 1, 3 and 10  $\mu$ g/g final measured concentrations of selo-L-methionine in test beakers with overlying seawater for 10 days. Two tests were conducted in static non-renewal exposure (one with adults and one with juveniles) and one test was conducted with juveniles and daily overly seawater renewal. The results from the static test using juveniles found a 10-day LC50 value of 1.6  $\mu$ g/l, a NOEC value of 0.84  $\mu$ g/l and a LOEC value of 2.0  $\mu$ g/l. The results from the static test using adults found a 10-day LC50 value of 7.6  $\mu$ g/l, a NOEC value of 1.0  $\mu$ g/l.

The results from the renewal sediment test using juveniles found a 10-day LC50 value of 6.3  $\mu$ g/l, a NOEC value of 1.7  $\mu$ g/l and a LOEC value of 5.0  $\mu$ g/l. Both juveniles and adults were sensitive to seleno-L-methionine, but daily renewal was found to reduce the toxicity of seleno-L-methionine spiked sediments to juveniles. This is potentially due to the lower concentrations of selenium in the overlying water in each renewal treatment (data not reported). Seleno-L-methionine was readily desorbed from the sediment into the overlying water column in concentrations that potentially contributed to observed toxicity. In addition, in a 10-day sediment test, sediment was collected from three sites in Lake Macquarie with high selenium concentrations ranging from 1.7  $\mu$ g/g to 6.8  $\mu$ g/g (dry weight). The concentrations of selenium had no significant effects on *Corophium* sp. survival. Hyne *et al.* (2002) concluded that seleno amino acids, particularly seleno-L-methionine are acutely toxic to *Corophium* sp.

**lliff et al. (2019)** investigated the effects of carbaryl on oyster reef community composition using slow-dissolving plaster blocks containing carbaryl and Travertine (limestone) tiles to simulate the hard-bottom microhabitat of an oyster reef. Additional plaster blocks without carbaryl were used as a procedural control. The dissolution blocks were replaced every three to five days. Sessile and epibenthic organisms were allowed to colonize the tiles for 28 days. After 28 days, the taxa colonized on the tiles were recorded *Americorophium* spp. was one of the most abundant and frequently observed invertebrates on the recruitment tiles, along with tube-forming serpulid worms *Hydroides* sp., eastern oyster *Crassostrea virginica*, ivory barnacles *Balanus ebureus* and gammaridean amphipods *Hourstonius laguna*. In the controls. This species was the only one in the study to show a significant decline in abundance in the carbaryl-treated blocks. A significant dissimilarity was observed between the carbaryl treatments and the controls amongst crustacean species, and it was suggested that this was primarily driven by the lack of *Americorophium* spp. in the carbaryl treatments. This indicated that *Americorophium* spp. was driving the shift in crustacean community



composition. Overall, carbaryl influenced oyster reef communities but the effects varied by species. Iliff *et al.* (2019) suggested the strong impact of carbaryl on *Americorophium* spp. measured in the study was because they build their soft tubes out of local sediments and organic materials.

Kirkpatrick et al. (2006) examined the behavioural effects of Corophium volutator, collected from Horse Island, Northern Ireland, to the lipophilic biocide Bioban, using a multispecies freshwater biomonitor (MFB). In laboratory conditions, Corophium volutator was exposed to sediment spiked with 56 mg/kg, 100 mg/kg, 121 mg/kg, and a control in a MFB for one hour. The percentage activity time, medium (sediment or overlying water) and activity type (locomotory activity or ventilation/ inactivity) were recorded. Bioban concentrations significantly affected the overall percentage activity duration of the tested amphipods. The percentage activity duration was significantly lower in *Corophium volutator* exposed to 56 mg/kg compared to 0 mg/kg, 100 mg/kg, and 121 mg/kg of Bioban. There was more Corophium volutator activity (both types) in the water column than in the spiked Bioban sediments, exhibiting a significant effect of the medium. As Bioban concentrations increased, there was an increase in the percentage of time Corophium volutator spent in locomotory activity compared to ventilation/inactivity. The evidence presented showed a rapid behavioural response to sublethal concentrations of Bioban. Kirkpatrick et al. (2006) concluded that the increase in locomotory activity exhibited in toxic sediment and nontoxic overlaying water showed avoidance and escape behaviour by Corophium volutator.

**Krang & Dahlstrom (2006)** investigated the effects of antifouling biocide medetomidine on pheromone induced male mate search behaviour in adult *Corophium volutator*, collected from Havstensfjord, Sweden, using a Y-maze choice test apparatus system. In laboratory conditions, male *Corophium volutator* individuals were exposed to 0 (control), 0.01 or 0.1 µg/l nominal concentrations of medetomidine in seawater for one day. Each exposure tank contained 12 male individuals. After exposure for one day, a single male individual from the three treatments was placed into a starting tank connected to a Y-shaped tube split into two arms, each leading to an arrival tank. One of the arrival tanks contained female odour from a connecting tank which contained sediment and 50 female *Corophium volutator*, the other arrival tank was empty of female odour. There was a continuous flow of seawater in the system. Each male had three hours to decide whether to stay in the starting tank or to crawl into one of the two arms, to determine male attraction towards the female odour. All other signals (acoustic or visual) were excluded. Each *Corophium volutator* male was only tested once, and the same aquaria were used for a week before the sediment and odour-producing



females were replaced. In a second round of experimental trials, using the same method and medetomidine concentrations, the end tank chosen by the test male was recorded. Exposure to medetomidine significantly affected the male *Corophium volutator* search behaviour. Males from the control treatment responded to female odour, as 55% of them left the starting tank and almost all of those moved towards the arrival tank with female odour. The distribution of males from the control was significantly different to males exposed to medetomidine concentrations. In the males from the 0.01 µg/l medetomidine exposure, 32% left the start tank (18 out of 56 individuals) and most of these crawled to the arrival tank with female odour (14 out of 18 individuals). In the males from the 0.1 µg/l medetomidine exposure, 16% left the start tank (9 out of 55 individuals) and these were observed to randomly select the end tank with or without the female odour. Krang & Dahlstrom (2006) concluded that medetomidine impairs the pheromone-induced mate search in *Corophium volutator* males, which could potentially delay mate location, increase predation risk, and affect population growth or recovery.

Mayor et al. (2008) examined the effects of five chemotherapeutic agents; copper, cypermethrin, oxytetracycline hydrochloride, azamethipos and emamectin benzoate on Corophium volutator, using laboratory-based whole sediment bioassays. These chemotherapeutic agents are the active ingredients in commercial treatments used to treat sea lice-infested salmon in Atlantic salmon fish farms. Corophium volutator was exposed to sediment samples (collected from an uncontaminated site in the Ythan estuary, Scotland) spiked with nominal concentrations of the five contaminants in a total of 210 mesocosms for 10 days. Nominal wet sediment concentrations included 30,170, 90,510, 181,020, 301,700 and 603,400 µg/kg of copper; 0, 0.1, 0.5, 5, 50 and 500 µg/kg of cypermethrin; 0, 10, 100, 1000, 10,000 and 100,000 µg/kg of oxytetracycline hydrochloride; 0, 10, 100, 1000, 10,000 and 50,000 µg/kg of azamethiphos; 0, 1, 10, 100, 1,000 and 10,000 µg/kg of emamectin benzoate. There were five replicates for each treatment, and 30 Corophium volutator individuals were added to each replicate. The percentage mortality was recorded. The concentration of each active ingredient had a significant effect on mortalities observed in each experiment, with the percentage mortality of *Corophium volutator* increasing with increasing contaminant concentration. Mayor et al. (2008) reported 10-day LC50 values of 193,326 µg/kg of copper, 5 µg/kg of cypermethrin, 414 µg/kg of oxytetracycline, 182 µg/kg of azamethiphos and 153 µg/kg of emamectin benzoate. Mortality was observed throughout the 10 days, but notably 100% mortality occurred within 24 hours at the highest concentrations of azamethiphos. In addition, figure 1 showed 100% mortality was also recorded in the highest



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concentrations of copper (603,400 µg/kg), emamectin benzoate (1000 µg/kg), azamethiphos (1000, 10,000, 50,000 µg/kg) and cypermethrin (500 µg/kg). Sublethal effects were observed as *Corophium volutator* swam erratically in copper exposures and rarely constructed burrows in high exposures. Overall, all the chemotherapeutic agents had toxic effects on *Corophium volutator*, but the evidence indicated that cypermethrin was the most toxic contaminant. In addition, Mayor *et al.* (2008) noted that *Corophium volutator* was able to tolerate higher copper concentrations than *Hediste diversicolor*, another marine species tested in the study. The full list of the percentage mortalities is shown in the evidence summary spreadsheet.

**Parlapiano** *et al.* (2021) examined the effects of three commercially used glyphosate-based herbicides; Roundup, Efesto and Taifun, on *Corophium insidiosum,* collected from Mar Piccolo of Taranti, using laboratory-based static toxicity tests. The study considered the effects of temperature on the toxicity of the herbicides to *Corophium insidiosum,* by conducting experiments at 20°C and 30°C. All experiments consisted of five concentrations of each herbicide, one control and three replicates per treatment. 20 individuals were added to each replicate.

In the Efesto experiment, juvenile *Corophium insidiosum* were exposed to 2, 4, 8, 16 and 32 mg/l nominal concentrations of Efesto at 20°C and 1, 2, 4, 8 and 16 mg/l nominal concentrations of Efesto at 30°C for 96 hours. The recorded 96-hour LC50 value was 7.94 mg/l in 20°C and 3.25 mg/l in 30°C. Significant *Corophium insidiosum* mortality was observed at 4 mg/l (at 20°C) and 2 mg/l (at 30°C).

In the Taifun experiment, juvenile *Corophium insidiosum* were exposed to 30, 60, 90, 120 and 150 mg/l nominal concentrations of Taifun at 20°C and 7.5, 15, 30, 60 and 120 mg/l nominal concentrations of Taifun at 30°C for 96 hours. The recorded 96-hour LC50 value was 59.04 mg/l in 20°C and 29.49 mg/l in 30°C. Significant *Corophium insidiosum* mortality was observed at the very high Taifun concentrations. In the Roundup experiment, juvenile *Corophium insidiosum* were exposed to 50, 100, 150, 200 and 250 mg/l nominal concentrations of Roundup at 20°C and 25, 50, 75, 100 and 125 mg/l nominal concentrations of Taifun at 30°C for 96 hours. The recorded 96-hour LC50 value was 111.17 mg/l in 20°C and 53.12 mg/l in 30°C. Significant *Corophium insidiosum* mortality was observed at the very high Roundup concentrations.

Overall, Parlapiano *et al.* (2021) reported significantly higher mean mortality in all concentration groups for all tested crustaceans, including *Corophium insidiosum* exposed to the three herbicides . In all experiments, there was a concentration-response relationship



observed in amphipod mortality. In addition, amongst the crustaceans tested in this study, juvenile *Corophium insidiosum* and *Tigriopus fulvus* nauplii were significantly more sensitive to each herbicide than the other test species (anostracan *Artemia franciscana* and isopod *Sphaeroma serratum*). Parlapiano *et al.* (2021) suggested that the different life stages did not affect the species' sensitivity. Efesto was the most toxic herbicide at both temperatures. In *Corophium insidiosum* Efesto was approximately seven times more toxic than Taifun and nearly twice as toxic as Roundup. In addition, the higher temperature exposure (30°C) increased the sensitivity of all tested crustaceans to the glyphosate herbicides.

**Perron et al. (2012)** investigated the effects of the pesticide triclosan exposure on adult *Ampelisca abdita*, collected from the Narrow River, Narragansett, in laboratory-based static-renewal water-only and whole sediment acute toxicity tests. In the water-only experiments, *Ampelisca abdita* was exposed to 10 μg/l, 30 μg/l, 100 μg/l and 300 μg/l of diluted triclosan in seawater, for 96 hours. Mortality was assessed daily. Results for this study found a 24-hour LC50 value of 187 μg/l and a 96-hour LC50 value of 73.4 μg/l triclosan. In the whole sediment experiment, *Ampelisca abdita* was exposed to 80 mg/kg to 800 mg/kg dry weight of triclosan spiked sediment for 7 and 10 days. Mortality was recorded at the end of exposure. Results found a 7-day LC50 and 10-day LC50 of 303 mg/kg and 260 mg/kg dry sediment weight, respectively. The authors concluded that *Ampelisca abdita* is sensitive to triclosan exposure, with similar toxicity values in seawater and sediment exposure. It was suggested that the amphipod *Ampelisca abdita* and mysid *Americamysis bahia* tested in this study, are some of the most sensitive species to triclosan, based on a lethality endpoint.

**Soto et al. (2000)** examined the sensitivity of juvenile *Ampelisca araucana* from the Chilean coast to organic and inorganic toxicants dissolved in sea water. The toxicants tested were the fungicide pentachlorophenol (PCP), potassium dichromate, copper (Cu) and herbicide 2,4-D. Each experiment had five treatments of each toxicant, four replicates, and five individuals. *Ampelisca araucana* juveniles were exposed to PCP concentrations 0.01, 0.1, 1, 10, 100 mg/l; 0.0125, 0.025, 0.05, 0.1 mg/l and 0.0625, 0.125, 0.25, 0.5, 1 mg/l for 48 hours. This experiment found a mean 48-hour LC50 value of 0.09 mg/l PCP. *Ampelisca araucana* juveniles were exposed to 50, 100, 200, 400, 800 mg/l of 2,4-D for 48 hours resulting in a mean 48-hour LC50 of 91.2 mg/l. *Ampelisca araucana* juveniles were exposed to 38.5, 55, 78.5, 112, 160 mg/l of potassium dichromate for 48 hours, resulting in 48-hour LC50 value of 56.89 mg/l. *Ampelisca araucana* juveniles were exposed to 62.5, 125, 250, 500, 1000 µg/l of Cu for 48 hours, resulting in found a 48-hours LC50 value of 305.5 µg/l. Soto *et al.* (2000) noted that early developmental stages are generally more sensitive to stress than adult



forms. Their results suggested that *Ampelisca araucana* juveniles were relatively sensitive to inorganic and organic chemicals, with juveniles less sensitive to metals and more sensitive to organic compounds.

**Tucca et al. (2014)** examined the toxicity of antiparasitic pesticides (emamectin benzoate, cypermethrin, and deltamethrin) on *Monocorophium insidiosum* (syn. *Corophium insidiosum*) using whole sediment bioassays. Specimens were exposed to sediment spiked with a range of five concentrations of each chemical for 10 days and mortality estimated. The 10-day LC50s were 890 µg/kg for emamectin benzoate, 57 µg/kg for cypermethrin and 7.8 µg/kg for deltamethrin. Tucca *et al.* (2014) noted that their 10-day LC50 for emamectin benzoate was higher than values reported for *Corophium volutator* of 183 µg/kg (Mayor *et al.*, 2008) or 193 µg/kg (SEPA, 1999, unseen). In contrast. their LC50 for cypermethrin was similar to the 10-day LC50 of 42 µg/kg reported by Millar (1999, unseen). Overall, deltamethrin was the most toxic antiparasitic tested. Sublethal antioxidant responses (GST and lipid peroxidation) were observed in short-term (2-day and 10-day) low concentration exposures to emamectin benzoate and cypermethrin, but not deltamethrin (Tucca *et al.*, 2014).

#### 5.2 Pharmaceuticals

Only two articles examined the effects of pharmaceuticals on the amphipods reviewed. Mayor *et al.* (2008) is summarized above (Section 5.1) and Gronlund *et al.* (2024) below.

**Gronlund et al. (2024)** examined the effects of pharmaceutical drugs diclofenac (a nonsteroidal anti-inflammatory drug) and citalopram (a selective serotonin reuptake inhibitor antidepressant) on *Corophium volutator* in laboratory-based sediment toxicity tests. *Corophium volutator* was collected from Herslev Beach and Lyndby Harbor, Denmark. Sediment collected from Herslev Beach was spiked with diclofenac sodium and/or citalopram dissolved in methanol. Experimental treatments exposed the amphipod for 10 days to nominal concentrations of diclofenac ranging from 0.1 to 100 µg/g (dry weight) or nominal concentrations of citalopram ranging from 0.1 to 200 µg/g (dry weight). A third experiment exposed the amphipod to a mixture of the pharmaceuticals, which included nominal concentration of citalopram. The constant concentration of citalopram was based on the LC50 value obtained in the single-compound experiment. Mortality was reported after 10 days of exposure. Gronlund *et al.* (2024) applied both time-weighted average assessments and the reduced general unified threshold model of survival (GUTS-RED model) to account for degradation and partitioning.



The results from the diclofenac exposure experiment found 10-day LC50 values of 11 µg/g (dry weight) based on time-weighted exposure and the GUTS-RED model. Measured start concentrations of diclofenac were 63% of the nominal concentrations and approximately 55.8% of the initial diclofenac had disappeared after 10 days and could not be recovered from the sediment or overlying water. The results found a 10-day LC50 value of 21.6 µg/g (dry weight), which was approximately 90% higher than the nominal concentrations. As the evidence showed diclofenac rapidly partitioning from the sediment into the water, Gronlund *et al.* (2024) stated that it is unlikely that diclofenac would pose a threat to sediment communities because the chemical properties of diclofenac make it more likely to be sufficient when assessing the environmental impact of diclofenac.

The results from the citalopram exposure experiment found 10-day LC50 values of between 64.9 and 97.3 µg/g (dry weight) based on time-weighted exposure and the GUTS-RED model. Measured start concentrations of citalopram were 104.9% of the nominal concentrations and on average 19.1% of the starting concentration of citalopram disappeared after 10 days. This suggested a slow dissipation rate of citalopram. Similar to exposure to diclofenac, *Corophium volutator* mortality increased drastically in a narrow range of concentrations. Gronlund *et al.* (2024) stated that citalopram is unlikely to directly pose a lethal threat to *Corophium volutator* community based on citalopram concentrations in water and sediment/water partitioning observed in the experiments. Overall results in the single-compound exposures, showed diclofenac was more toxic than citalopram to *Corophium volutator*. Results from the mixture study found a synergistic effect, meaning the combined impact of the contaminants was greater than expected (Gronlund *et al.* 2024).

## 5.3 Polychlorinated biphenyls (PCBs)

Polychlorinated biphenyls (PCBs) were examined in only two articles reviewed. Reish (1993) is summarized above (Section 4.1) and Ho *et al.* (1997) below.

**Ho et al. (1997)** examined the toxicity of the PCB mixtures Aroclor 1242, Aroclor 1254 and bis(2-ethylhexyl) phthalate on *Ampelisca abdita*, through laboratory-based toxicity tests. The study aimed to determine the toxic components in the pore water from New Bedford Harbor (NBH) sediments, a site known to be heavily contaminated with PCBs, PAHs, metals, polychlorinated dibenzofurans, and polychlorinated dibenzo-p-dioxins. In the first experiment, *Ampelisca abdita* was exposed to five unidentified concentrations of the PCB mixtures



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Aroclor 1242 and 1254 starting at 500 ppb, with each subsequent concentration following a 50% serial dilution. The PCB concentrations were renewed after 48 hours, and the total exposure duration was 96 hours. Aroclor 1242 was more toxic to the tested amphipod than Aroclor 1254. The 48-hour LC50 and 96-hour LC50 values for Aroclor 1242 were 21 ppb and 10 ppb, respectively, and the 48-hour LC50 and 96-hour LC50 values for Aroclor 1254 were 98 ppb and 40 ppb, respectively. The second experiment exposed *Ampelisca abdita* to 12, 37, 111, 333, 1,000 ppb measured concentrations of phthalic acid, bis(2-ethylhexyl) phthalate for 96 hours and concentrations were renewed after 48 hours. The resultant 48-hour LC50 was above 1,000 ppb of phthalate for *Ampelisca abdita*. This suggested that the phthalate was not toxic for amphipods in the samples studied. Ho *et al.* (1997) concluded that PCBs were likely to be the source of chronic and reproductive toxicity in the sediment studied.

### 5.4 Phthalates

Phthalates (phthalate esters) were examined by only three articles. Ho *et al.* (1997) is summarised above (Section 5.3) and Tagatz *et al.* (1983) below, while Tagatz & Stanley (1987; cited from ECOTOX) was unseen.

Tagatz et al. (1983) examined the effects of di-n-butyl phthalate (DBP) on field and laboratory benthic communities, which included the amphipod Corophium acherusicum. The field study was conducted in Santa Rosa Sound, Florida, using sand filled field aguaria that were colonized by naturally occurring communities. The laboratory study used an aquarium that was colonized by planktonic larvae entrained in a continuous supply of unfiltered seawater from Santa Rose Sound. Following eight weeks of colonization, eight aquaria for each treatment (field and laboratory colonised communities) were exposed for two weeks to a control and DBP measured at concentrations of 0.044, 0.34 and 3.7 mg/l (laboratorycolonized) or 0.036, 0.45 and 3.8 mg/l (field-colonized). Pure DBP was dissolved in a stock solution of 60% acetone and 40% distilled water. Field communities were moved to the laboratory for exposure. The resultant community structure, in both field and laboratory communities, was altered in the 3.7 mg/I DBP treatments, with significantly fewer individuals and species of animals in the highest concentration of DBP compared to the control and lower DBP concentration treatments. *Corophium acherusicum* only colonized the laboratory aquaria but was one of the dominant species found. In the total of 2,331 marine animals identified in this community, there were 177 Corophium acherusicum. The number of Corophium acherusicum significantly decreased to eight individuals in the 0.34 mg/I DBP treatment. There were zero Corophium acherusicum individuals recorded in the 3.7 mg/I DBP



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treatment. The density of individuals and numbers of species was not affected by 0.044 mg/l DBP, with 75 *Corophium acherusicum* individuals identified in this concentration and 94 individuals in the control. This evidence shows that *Corophium acherusicum* in laboratory colonized communities were significantly affected by as low a concentration as 0.34 mg/l DBP. Tagatz *et al.* (1983) reported that the presence of acetone could have influenced bioavailability and toxicity, but there was no data to support this.

### 5.5 Synthetics (other)

The 'synthetics (other)' category includes a range of chemicals that do not fit into other categories conveniently. Hence, several of the chemicals included under this category only appeared in one or two studies. At total of eight results were obtained from the six studies reported the effects of 'Synthetics (other)' on selected amphipod species. Lera *et al.* (2008) and Picone *et al.* (2008) are summarized in above (Section 4.1). However, Thursby & Berry (1987; cited from ECOTOX) and Harris & Morgan (1984; cited from ECOTOX) were unseen. The remaining three articles are summarized below.

**Hester et al. (1991)** examined the effects of Arosurf MSF (monomolecular surface film) on an unidentified amphipod and other marine organisms, through laboratory-based acute static toxicity tests. Arosurf MSF is a surfactant used to control mosquito larvae by creating an oily film over the water surface that prevents the larvae adhering to the surface to breathe and interferes with emergence by adults. In the study, organisms were exposed to a maximum dosage of 47 ml/m<sup>2</sup> of Arosurf MSF in beakers containing 400 ml of water for 96 hours. There was 0% adjusted mortality found in *Gammarus* sp. The results indicated that Arosurf MSF had no acute impact on the life stages of any tested organism, demonstrating no toxicity at this concentration.

**Fastelli & Renzi (2019)** examined the effects of different sunscreens on three marine species of ecological significance, including *Corophium orientalis*. Two different sunscreen formulas were tested, a chemical-based UV filter (reported as Bis-Ethylhexyloxyphenol methoxyphenyl triazine; Butyl methoxydibenzoylmethane; Ethylhexyl methoxycinnamate; Ethylhexyl salicylate) and a physical sunscreen type based on metal oxides in their nanoform as a UV filter (reported as n-TiO2; n-ZnO). In laboratory conditions, *Corophium orientalis* was exposed to geometric dilutions of a sunscreen and water ratio, which started at 100 μl/l. Experiments were conducted under two different salinities; standard optimal salinity (3.5 ppt) and osmotic stress (4.0 ppt). The survival rate for the amphipod was recorded after 96 hours. Results for *Corophium orientalis* under 3.5 ppt salinity found a 96-hour LC50 of 96 μl/l for the



chemical-based sunscreen and a 96-hour LC50 over the maximum concentration tested (100  $\mu$ I/I) for the physical-based sunscreen type. The chemical-based sunscreen had significantly lower toxicity to *Corophium orientalis* than the physical-based sunscreen type at higher salinity. Under salinity stress (4.0 ppt), increased toxicity was found in both sunscreen types. A significant effect between sunscreen types and salinity stress was recorded. The 96-hour LC50 in 4.0 ppt salinity was 82  $\mu$ I/I for chemical-based sunscreen and 87  $\mu$ I/I for physical-based sunscreen. The study also conducted positive control tests, which directly exposed the tested amphipod to the standard toxicant cadmium chloride for 96 hours and found an LC50 range of 1.56 to 4.38 mg/I of cadmium.

Fastelli & Renzi (2019) concluded that chemical-based and physical-based sunscreen types can exert different ecotoxicological responses on the tested species, suggesting that sensitivity toward nanoparticles in personal care products could be very different amongst different marine taxa. The evidence also suggested that salinity stress could affect changing ecotoxicological responses, as under osmotic stress the toxicity of the sunscreens was amplified.

Rico-Rico et al. (2009) examined the toxicity of the surfactant C12-2-LAS (2-(psulphophenyl)-dodecane) to Corophium volutator in both water and sediment bioassays. In the water-only assays, Corophium was exposed to a range of nominal concentrations (0.2 to 1.6 mg/I LAS) for 2, 3 and 5 days and mortality assessed. The 2-day LC50 value was 1.00 mg/I LAS, the 3-day LC50 value was 0.19 mg/I, and the 5-day LC50 was 0.73 mg/I LAS based on the actual concentration. Two sets of sediment assays were conducted. In the first set, sediment from the Oesterput were spiked with 50 and 500 mg/kg LAS and Corophium exposed for 1, 3, 5, 7 and 10 days. Exposure to 50 mg/kg was not significantly different from controls, while mortality increased to ca 100% after 10 days at 500 mg/kg (based on Figure 4; Rico-Rico et al., 2009). Most (60 to 80%) of the final mortality occurred within three days of exposure to 500 mg/kg. In the second experiment sediment from Oesterput and Cadiz was spiked with 60 and 600 mg/kg LAS and Corophium exposed for 5 days. The resultant 5-day LC50s were 295 and 162 mg/kg LAS in sediment (actual concentration) and 0.47 and 1.24 LAS in pore water (actual concentration based on sorption) in Oesterput and Cadiz sediment, respectively. Rico-Rico et al. (2009) concluded that water-only LC50s were similar to pore water LC50s.



# 6 Other substances

A total of 15 results (ranked 'worst-case' mortalities) were obtained from eight articles that examined the effects of 'Inorganic chemicals' on amphipod species.

**Caldwell (1975)** investigated the effects of hydrogen sulphide (as sodium sulphide) on *Corophium salmonis* collected from Yaquina Bay, through acute toxicity tests. In laboratory conditions, *Corophium salmonis* was exposed to 1.0, 3.3, and 10.0 mg/l of hydrogen sulphide in seawater in exposure containers for 96 hours. There were 10 *Corophium salmonis* per concentration and mortality was recorded every 24 hours. Results found the 24-hour LC50 value was 1.4 mg/l of hydrogen sulphide, and the 48-hour LC50 and 96-hour LC50 value was less than 1.0 mg/l of hydrogen sulphide. The author concluded that *Corophium salmonis* are unable or barely able to tolerate 1.0 mg/l of hydrogen sulphide during continuous exposure for multiple days.

**Ferretti** *et al.* (2004) conducted acute sediment toxicity tests using the amphipod *Ampelisca abdita*, at six laboratories in the United States. Each laboratory used five unidentified concentrations of potassium chloride (KCI), from a stock solution of 75,000 mg/l, as a reference toxicant in water-only exposure tests with *Ampelisca abdita*. Results from the reference toxicant tests at each laboratory found 96-hour LC50 values of 1,080 mg/l of KCI, 605 mg/l of KCI, 1,370 mg/l of KCI, 819 mg/l of KCI, 798 mg/l of KCI and 727 mg/l of KCI. The authors noted that these results compare favourably to water-only reference toxicant data published in other literature.

**Ho et al. (1999a)** studied the effects of ammonia uptake by *Ulva lactuca* and considered the ammonia toxicity to the marine amphipod *Ampelisca abdita* and mysids. In laboratory conditions, acute static assays exposed juvenile amphipods to nominal concentrations of 100 mg/l of ammonia in seawater for 48 hours in the presence and absence of *Ulva lactuca*. *Ulva lactuca* reduced ammonia concentrations from 75 mg/l to 17 mg/l in 8 hours, which also reduced amphipod mortality from 75% to 20%. The experiment suggested that *Ulva lactuca* uptake of ammonia removed all toxicity for the amphipods.

**Kater** *et al.* **(2006)** examined the effects of total ionized ammonium (NH<sub>4</sub><sup>+</sup>) and un-ionized ammonia (NH<sub>3</sub>) on *Corophium volutator*, collected from Oesterput, The Netherlands. The study conducted a series of laboratory-based sediment and water-only toxicity tests at different controlled pH levels to assess the influence of pH levels on toxicity. In the water-only



toxicity tests, *Corophium volutator* was exposed to total ammonium chloride dissolved in seawater at varied nominal concentrations between 0 to 100 mg/l at low pH and 0 to 45 mg/l at high pH for 72 hours. The experiments were conducted in glass beakers with no sediment and with twenty randomly selected *Corophium volutator* in each beaker. There were three replicates used for each concentration. The number of surviving *Corophium volutator* was recorded after 72 hours. The water-only test was carried out eight times at different pH levels (8.2, 8.3, 8.4, 8.5, 8.6, 8.7, 8.8, 8.9, 9 and 9.1). The results found a significant decrease in 72-hour LC50 values as the pH increased. The 10-day LC50 varied between 10 and 85 mg/l (extracted from Figure 1; Kater *et al.*, 2006). The specific LC50 values for all experiments were reported in Figures 1 and could not be extracted.

In the sediment toxicity tests, *Corophium volutator* was exposed to total ammonium concentrations dissolved in seawater at varied nominal concentrations between 0 to 320 mg/l at low pH and 0 to 100 mg/l at high pH for 10 days. The experiments were conducted in glass beakers, which contained 600 ml of contaminated seawater and 200 ml of sediment collected from Oesterput, there were twenty randomly selected *Corophium volutator* in each beaker. There were three replicates used for each concentration. The number of surviving *Corophium volutator* was recorded after 10 days. The average measured total ammonium concentrations after 10 days varied between 1.7 and 5.3 mg/l. The sediment test was carried out 12 times; six in the winter and six in the summer, under varied pH levels (8.0, 8.1, 8.2, 8.3, 8.4, 8.5, 8.6, 8.7 and 8.8). The results found a significant decrease in 10-day LC50 values as the pH increased. The 10-day LC50 values varied between 30 and 80 mg/l (based on overlying water nominal concentrations), depending on pH levels. The evidence shows that LC50 values were higher in the sediment tests compared to the water-only tests, suggesting the *Corophium volutator* experiences stress in the water only experiments. The specific LC50 values for all experiments were reported in Figures 2 and could not be extracted.

The sediment experiments with pH above 8.3 were used to test un-ionized ammonia toxicity. The NH<sub>3</sub> concentrations, which ranged from 0.5 to 9.6 mg/l, were determined from each total ammonium concentration in the sediment experiments. The results found a 72-hour LC50 value of 3.1 mg/l. The LC50 value was used to estimate expected LC50 values for total ammonium at each pH level, assuming toxicity is only caused by ammonia. The results suggest that at a pH level above 8.3, all LC50 values align with the confidence range of the expected toxicity. However, toxicity was higher than expected at lower pH levels, suggesting combined toxicity of ammonium and un-ionized ammonia at pH levels below pH 8.3. Figure 3 (see Kater *et al.*, 2006) shows these expected values, along with LC50 results from pH-



controlled sediment experiments. Kater *et al.* (2006) concluded that water quality criteria for total ammonium concentrations in overlying water, in sediment bioassays with *Corophium volutator* should be pH dependent.

**Re et al. (2007)** examined the suitability of *Corophium multisetosum* as a bioassay for water quality when compared to other standard bioassay species. The species studied were exposed to 0, 3.2, 6.25, 12.5, 17.5, 20 and 25% of boiling cork effluent and 0, 0.8, 1.6, 3.2, 6.25, and 12.5% iron filings lixiviates for 96 hours. (lixiviates refer to the soluble fraction washed out of iron filings). *Corophium* experienced >80% mortality (based on Figure 1) in 25% boiling cork effluent, with a 96-hour LC50 of 15.74%. *Gammarus chevreuxi* was more sensitive with a 96-hour LC50 of 14.91 boiling cork effluent, and 100% mortality in 25% effluent. The tested species were more sensitive to the iron filing lixiviates. *Corophium* experienced >90% mortality at 12.5% lixiviates with a 96-hour LC50 of 1.56% lixiviates. *Corophium multisetosum* was also exposed to 10 separate sediment samples from the Ria de Aveiro, Portugal in 10-day assays. Mortality were only significantly different from the control in one sample, which Re *et al.* (2007) suggested showed the signs of organic enrichment.

**Smit** *et al.* (2008) investigated the effects of hydrogen peroxide, used to disinfect ballast water, on *Corophium volutator* and other marine species. In laboratory conditions, *Corophium volutator* was exposed to water contaminated with 10 to 160 mg/l of hydrogen peroxide for 96 hours. The number of mobile individuals was recorded after 24, 48, 72 and 96 hours of exposure. The study used time-dependent dose-response curves to gain time-dependent species-sensitivity distributions (SSDs), which describe a combination of time and effect concentration which affects a certain percentage of marine species. For all species tested, the effect levels (fraction immobilised) increased as exposure time and duration increased. At higher hydrogen peroxide concentrations, there was a higher fraction of immobilised *Corophium volutator*. Results observed a 24-hour EC50 of 611 mg/l and a 96-hour EC50 of 46 mg/l. *Corophium volutator* had a relatively low sensitivity to hydrogen peroxide compared to *Artemia salina, Brachionus plicatilis, Dunaliella teriolecta*, and *Skeletonema costatum,* which were the other species tested in the study.



# 7 Contaminated sediment

A total of 30 results (ranked 'worst-case' mortalities) were obtained from 19 articles that examined the effects of 'contaminated sediment' on amphipod species. *Corophium* spp. accounted for most of the contaminated sediment results (86.7%), followed by *Ampelisca* spp. (6.7% of results). As stated above, the contaminated sediment articles were not used in the sensitivity assessment of the separate contaminant types because contaminants may act synergistically or antagonistically in mixtures and it was not obvious which one or more the contaminants caused the reported toxicity. They are summarized below for information.

Birch et al. (2008) examined the effect of contaminated sediments from Sydney Harbour, Australia, on the survival of amphipod Corophium colo in laboratory-based whole-sediment toxicity tests. Sediment samples were collected from multiple sites across the estuary, which were contaminated with heavy metals, organochlorine pesticides, hexachlorobenzene (HCB), total polychlorinated biphenyl (PCBs), polycyclic aromatic hydrocarbons (PAHs). The mean measured concentrations in the sediment samples included 210 mg/kg of copper, 390 mg/kg of lead, 900 mg/kg of zinc, 3,100 µg/kg of benzo(b)fluoranthene, 4,300 µg/kg of fluoranthene, and 5,100 µg/kg of pyrene. The total mean concentration of PCBs in the sediment was 40 µg/kg, with mean measured concentrations including 150 µg/kg total DDT, 2.8 µg/kg of chlordane, 7.2 µg/kg of dieldrin and 37 µg/kg of hexachlorobenzene. Corophium colo was exposed to the contaminated sediment for 10 days, and survival rates were compared to uncontaminated control sediments. The results found that the toxicity increased with increasing concentration of contaminants. Mortality in amphipods was unspecified. The amphipod survival was lower than predicted by sediment quality guidelines (SQGs), with mortality increased in areas with high chemical contamination. However, the species was less sensitive compared to sea urchin, Heliocidaris tuberculata, another tested organism. The effect of contaminated sediment on sea urchin fertilization had a stronger toxic response than the survival test for Corophium volutator. Birch et al. (2008) concluded that the sediments in Sydney Harbour were toxic, especially to amphipod survival and sea urchin development and fertilization.

**Botwe et al. (2017)** investigated the effect of contaminated sediments from Tema Harbour, Ghana on *Corophium volutator* using a whole-sediment bioassay. Tema Harbour was contaminated due to maritime operations and industrial activities, leading to high levels of PAHs, pesticides, and heavy metals in the sediment. Sediment samples were collected from 30 stations in the Tema Harbour using a stainless steel grab, these grab samples were then



combined into five composite samples. *Corophium volutator* was collected from an uncontaminated site in the Eastern Scheldt, The Netherlands and exposed to the contaminated sediment for 10 days in laboratory conditions. There were twenty *Corophium volutator* per replicate and five replicates per sediment sample and control. Mortality and metal bioaccumulation were recorded at the end of the experiment.

The measured metal concentrations in the sediment samples varied across locations, with cadmium (Cd) concentration ranging from 0.07 mg/kg to 1.16 mg/kg, lead (Pb) from 24.9 mg/kg to 102 mg/kg, chromium (Cr) from 50.1 mg/kg to 80.3 mg/kg, nickel (Ni) from 17.4 mg/kg to 27.7 mg/kg, copper (Cu) from 33.4 mg/kg to 210 mg/kg, zinc (Zn) from 98 mg/kg to 730 mg/kg, and arsenic (As) from 7.9 mg/kg to 14.2 mg/kg.

They reported significantly higher mortalities of *Corophium volutator* in the Tema Harbour sediments compared to the control, with mean percentage mortalities of 29%, 38%, 77%, 98% and 99% across the five sites. The highest mortalities were found in the Canoe Basin (CB) site (98%) that had the highest Cd concentrations and in the Inner Fishing Harbour (IFH) site (99%) that had the highest Cu and As concentrations. A statistically significant correlation was found between *Corophium volutator* mortality and Cd levels in the sediment. In addition, no burrowing activity was observed during the experiment in the CB or IFH sediments as *Corophium volutator* was avoiding the toxic sediment and staying in the water column. Botwe *et al.* (2017) concluded that Tema Harbour sediment was toxic and hazardous to *Corophium volutator*. The study found that *Corophium volutator* was more sensitive to sediment toxicity compared to *Hediste diversicolor*, another tested marine organism.

**Burgeot et al. (2017)** examined the effect of sediment contamination on *Corophium arenarium* using a sediment toxicity bioassay conducted in four locations: the Wadden Sea, Cartagena, the Seine estuary, and Iceland. The study exposed adult *Corophium* to sediment samples under static conditions for 10 days. The net percentage of mortality in undiluted sediment was recorded as 12.4% in the Wadden Sea and 10.7% in Cartagena. According to the USEPA (1998) guidelines, sediment toxicity is classified as significant at 20% mortality or higher; therefore, following these guidelines Burgeot *et al.* (2017), concluded that none of the sites represented toxic sediment conditions. Additionally, EC20 and EC50 thresholds were not reached in any of the four test sediments, further confirming low toxicity. Burgeot *et al.* (2017) suggested that *Corophium* mortality was not significantly impacted by sediment contamination at these locations, but variations in physico-chemical properties of the sediments could influence bioavailability and exposure.



Casado-Martinez et al. (2007) investigated the effect of dredged sediments from Spanish ports on adult amphipod species Corophium volutator and Ampelisca brevicornis, through laboratory-based sediment toxicity bioassays. Ampelisca brevicornis individuals were collected from an uncontaminated site in the Bay of Cadiz, and Corophium volutator individuals were collected from an uncontaminated site in Oesterput, Netherlands. The study exposed the amphipods to 22 sediment samples collected from different Spanish harbours for 10 days. There were twenty amphipod individuals added to each test chamber and three replicates for each sediment sample. The sediment samples were characterized for heavy metal concentrations (including cadmium, lead, copper, zinc, and chromium), PCB congeners and polycyclic aromatic hydrocarbons (including acenaphtylene, acenaphtene, anthracene, benz(a)anthracene, benz(a)pyrene, chrysene, dibenz(a,h)anthracene, phenanthrene, fluoranthene, fluorene, naphthalene, and pyrene). The sediments were classified according to Spanish regulatory thresholds, falling into three contamination categories. Both amphipod species exhibited high mortality when exposed to the contaminated sediment. There were 17 samples (sediment toxicity of 77%) in the Ampelisca brevicornis tests and 16 samples (sediment toxicity of 72%) in the Corophium volutator tests classified as toxic sediment. Both amphipod species had significantly different mortality from the control. The mean mortality of Corophium volutator ranged from 9.50 to 64.5%, and the mean mortality of Ampelisca *brevicornis* ranged from 16.7% to 78.9% (taken from Table 3). Casado-Martinez *et al.* (2007) found that Ampelisca brevicornis mortality correlated more with metal contamination, while Corophium volutator mortality correlated more with organic micro-pollutants (e.g. PCBs and PAHs). Overall, both amphipod species showed a similar toxic response to high contamination levels, Corophium volutator was more sensitive to the lower toxicity levels.

**Costa et al. (2016)** examined the effects of contaminated sediment from Mar Piccolo of Taranto on marine organisms including *Corophium insidiosum*. Mar Piccolo of Taranto is an enclosed basin with two shelves, the 'first inlet' and 'second inlet.' Sediment samples were collected from eight stations in the first inlet of Mar Piccolo (1A, 1D, 1I, 1C, 1E, 1H, 1F, 1G) and three in the second inlet (2A, 2B, 2C). Chemical analysis revealed sediment samples were strongly contaminated with a range of heavy metals, PAHs, and PCBs. Some of the concentrations of contaminants exceeded sediment quality guidelines, particularly in the first inlet, which showed very high PAH concentrations. Heavy metal concentrations were very high, except for copper and zinc. Overall, the sediment samples from the first inlet were more contaminated than those from the second inlet. In laboratory conditions, *Corophium insidiosum* collected from an uncontaminated site in the Gulf of Taranti were exposed to

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contaminated sediment samples for 10 days. In each experiment, 20 randomly selected young adults were added to a glass beaker containing one sediment sample and filtered seawater. The number of surviving amphipods was recorded after 10 days. Only sediment samples from stations 1D, 1I, 1C and 1E resulted in mortality higher than the test validity threshold (15% mortality in the control). The percentage mortality was less than 30% for all sediment samples, therefore an LC50 value could not be determined, and the results were not significant. Costa *et al.* (2016) concluded that despite chemical results confirming sediment from Mar Piccolo was contaminated there was an unexpectedly low toxic effect for all organisms tested in the study. It was suggested that the contaminants in the sediment are poorly bioavailable to *Corophium insidiosum* resulting in low toxicity and mortality.

Grant & Briggs (2002) examined the effects of sediment contaminated by drill cuttings from the North West Hutton oil platform in the North Sea on marine benthic organisms, including Corophium volutator, using laboratory-based sediment toxicity bioassays. Sediment samples were collected from 11 sites 100 m from the platform to 10,000 m from the platform along two parallel transects 10 m apart (labelled A and B). Sediment was also collected at one or more depth intervals; 5 to 10 cm, 10 to 15 cm and 15 to 20 cm. Sites were 100A, 100B, 200A, 200B, 300A, 300B, 400A, 400B, 500A, 500B, 600A, 600B, 800A, 800B, 1,200A, 1,200B, 2,500A, 2,500B, 5,000A, 5,000B, 7,500A, 7,500B, 10,000A and 10,000B. The sediment samples were analysed for contaminant concentrations and found a mixture of total hydrocarbon and seven heavy metals (Ba, Cr, Cu, Fe, Mn, Pb and Zn). The concentrations of hydrocarbon strongly correlated with the most toxic metals (Cu, Pb and Zn). Grant & Briggs (2002) noted that this strong correlation would make it difficult to distinguish between toxicity caused by metals and toxicity caused by hydrocarbons. Corophium volutator and control sediment was collected from Burgh Castle, Norfolk. In a pilot test, Corophium volutator was exposed to surface sediment from 19 sites and one subsurface sample in glass beakers with artificial seawater for 10 days. One replicate and 15 individuals per site. Results from the pilot test found sediment samples from sites closer to the oil platform (sites 100B, 300A, 300B, 400A, 400B and 500B) had significantly lower percentage survival than in the controls. Sediment from sites at 100 m and 300 m caused 100% mortality. There was also significantly high mortality observed in the sediment from site 2,500B in the pilot, but this was not significant in the main test with two duplicates, so the high mortality observed is assumed to be caused by other factors and not hydrocarbon toxicity.

In the main test, contaminated sediment samples from some key sites were diluted with clean sediment; sediment from 100 m had 1% and 3% dilutions, sediment from 200 m to 600 m



had 10% dilutions and sediment from 800 m outwards had 50% dilutions. *Corophium volutator* was exposed to the diluted sediment in glass beakers with artificial seawater for 10 days. There were two replicates and 20 individuals per replicate. Grant & Briggs (2002) found significant mortality from sites at 100 m to 600B, suggesting that sediment from sites closer to the oil platform were more toxic. However, a 25% dilution from site 400A and 10% dilution from site 200B made it non-toxic, as there was no significant difference in percentage survival from the control. Sediment from sites 100 m after 3% dilution caused 100% mortality.

Overall, contaminated sediment from the North West Hutton cutting pile was very toxic to *Corophium volutator*. Grant & Biggs (2002) concluded that the acute sensitivity of *Corophium volutator* to contaminated sediment from North West Hutton was higher than *Arenicola marina*, another species tested in this study. The evidence presented did not indicate which contaminant was responsible for the observed toxicity. However, the main test with dilutions found a weak relationship between *Corophium volutator* mortality and metal concentrations, which could suggest toxicity was unlikely to be due to metals.

Guerra et al. (2009) examined the effects of channel dredging, carried out in the Pialassa Baiona lagoon, Italy, using laboratory-based sediment toxicity tests on Corophium insidiosum and a before-after-control-impact (BACI) approach. The lagoon is exposed to industrial, urban, and agricultural wastewater, and previous literature has noted high level of mercury (Hg), polycyclic aromatic hydrocarbons (PAHs) and synthetic polymers in sediments. Sediment core samples were collected from 12 sites (impacted and non-impacted). There were six impacted sites; three in the dredged Baccarini channel (described as BAC 1, 2 and 3) and three in an adjacent pond (described as POL 1, POL 2 and VEN 5); and there were six non-impacted control sites; three sites in a channel (described as TBF 1, 3 and 4) and three sites in the pond (described as RIS 1, 2 and 3). Four replicate sample sediments were collected from each site before (September 2004) and after (September 2005) dredging. The concentrations of copper, cadmium, chromium, nickel, lead, and mercury in the sediment were determined. The heavy metal concentrations were plotted in Figure 3, and this shows increased concentrations of the metals, but Cd was undetectable in most cases. The metal concentrations fell within the expected ranges found in the Adriatic Sea, except for Hg, which ranged from 0.37 to 5.51 mg/kg and was well above background Hg concentrations found in the Mediterranean, Adriatic, and Ionian Sea. Corophium insidiosum and native sediment (used as a negative control) were collected from uncontaminated Valli di Comacchio Lagoon and twenty individuals were exposed to each sediment sample in glass beakers containing overlying seawater for 10 days. The results found 73 to 100% mean survival of Corophium



*insidiosum* in tested sediments across all sites and 40 to 100% survival for single beakers. This suggested that the sediments did have some effect on *Corophium insidiosum* survival but only two sites (POL 3 and TBF 4 before dredging) had a significant difference in survival from the negative control. There was no effect of dredging on sediment toxicity to *Corophium insidiosum* found in the study, as statistical tests revealed no significant difference when comparing the interaction between time and a putatively impacted site and the interaction between time and control site. If dredging had an effect the impacted sites would change over time and be different from patterns seen in the control sites. Guerra *et al.* (2009) concluded that the percentage survival of *Corophium insidiosum* was not correlated with any measured metal concentration or sediment properties. The two sites (POL 3 and TBF 4 before dredging), which were classified as toxic and had a significant effect on *Corophium insidiosum* survival, had average metal concentrations. Therefore, toxicity could have been caused by some other unidentified contaminant.

Macken et al. (2008) examined the sediment toxicity of marine sediments from three sites in Northern Ireland on *Corophium volutator*, through an acute whole sediment toxicity test and an ammonium chloride reference toxicant test. Sediment was collected from North Bull Lagoon (BL), Alexandra Basin (AB) and Dunmore East (DE) Harbour. Previous literature suggested that there are high levels of contamination by a variety of contaminants in the Alexandra Basin and high levels of organotin levels in Dunmore East Harbour sediment. Bull Lagoon was used as a control, and although it was found not to be a true control, it was still used as a control as it was the source of Corophium volutator. The study conducted a chemical analysis for heavy metals (mercury, aluminium, lithium, arsenic, cadmium, chromium, copper, lead, nickel, zinc) and organic contaminants (PAHs, PCBs, organochlorine pesticides (OCPs), TBT, DBT, MBT) in the contaminated sediment and this revealed varying contaminant levels across the three Irish sites (see Table 2; Macken et al., 2008). Corophium volutator individuals were exposed to the sediment in test vessels for 10 days. There were three replicates used per treatment and ten individual Corophium volutator in each. The test vessels were observed to make sure all amphipods burrowed into the sediment. Corophium volutator survival was recorded. BL sediments were assessed in March and November 2005 to confirm sediment suitability to use as control and to verify the amphipods available all year round. There was no significant mortality or temporal variation in toxicity of five different sampling occasions of the BL. A significant difference was found in the percentage mortality of Corophium volutator exposed to DE (approximately between 40 to 50% mortality) and AB sediments (60% mortality) compared to the control (BL) sediments



(evidence taken from Figure 6; Macken *et al.*, 2008). Alexandra Basin (AB) sediment was the most toxic. There was an increase in *Corophium volutator* individuals re-emerging from the AB sediment; this indicates a chemical avoidance behaviour. A 72-hour ammonium chloride reference toxicant test was also carried out to assess the sensitivity of the tests and determined a 72-hour LC50 value of 210.4 mg ammonium/l for *Corophium volutator*.

Matthiessen et al. (1998) examined the toxicity of contaminated sediment from the River Tyne Estuary, Northeast England on multiple marine organisms including *Corophium* volutator and Corophium arenarium through laboratory-based static whole sediment tests. Sediment samples were collected from six sites along the intertidal shoreline of the Tyne Estuary. Five sites were in the industrialised section of the estuary (Jarrow Slake, St Anthony's, Parish Quay, Ouse Burn, River Team) and one site was upstream from the main industrial wastewater inputs (Ryton). There were five sub-stations at the River Team site (River Team 1 to 5). Adult Corophium volutator (collected from Sandwich Bay, Kent) and Corophium arenarium (collected from Shoeburyness Sands, Essex) were exposed to the contaminated sediment in glass beakers with overlying seawater for 10 days. There were 2 to 3 replicates used and twenty amphipods per replicate. The distribution of heavy metals (cadmium, mercury, chromium, copper, nickel, lead, zinc), tributyltin, total hydrocarbons, total polychlorinated biphenyls, DDE and hexachlorobenzene in the sediment were analysed. . The highest contamination levels were observed in the River Team sub-stations 1 and 2, and the high contaminant levels in the River Team samples were broadly mirrored with the Corophium spp. toxicity data found in the study. The percentage mortality ranged from less than 10% to more than 70% mortality in Corophium volutator and ranged from 17% to 70% for Corophium arenarium between all River Tyne Estuary sites (taken from Figure 3; Matthiessen et al., 1998). In addition, mortality results from the River Team samples varied substantially between replicates. For example, there was 17% mortality recorded in Corophium arenarium in the River Team 5 sample, but 70% mortality recorded in the River Team sample 2. A burrowing sublethal response was observed in Corophium volutator, and this was less variable between the River Team replicates (Matthiessen et al., 1998).

**McCready** *et al.* (2005) examined the effects of contaminated sediment from Sydney Harbour, Australia and surrounding coastal lakes and estuaries south of Sydney on the survival and reburial of *Corophium colo*. A total of103 sediment samples collected .<u>collected</u>. The sediment samples were analysed for a range of contaminants including; 12 metals, 21 organochlorine pesticides, seven polychlorinated biphenyl Aroclors, 24 polynuclear aromatic



hydrocarbons, total organic carbon, and ammonia. Many samples exceeded established sediment quality guidelines, with up to 16 effects range-median (ERM) thresholds exceeded.

In laboratory conditions, adult and adolescent *Corophium colo* were exposed to the contaminated sediment samples and a negative control sample in test chambers with overlying water for 10 days. There were five replicates, and 16 to 20 individuals were added to each replicate. Mortality and individual length were recorded after 10 days. In addition to the survival tests, a reburial test was also conducted for 33 sediment samples; this test examined the ability of individuals to rebury in clean control sediment for 24 hours after exposure to contaminated sediment.

Their results classified 15 sediment samples (out of 103) as toxic compared to negative controls, with 11 of those classed as 'highly toxic' due to the mean survival of *Corophium colo* being less than 80% of the control and four classified as 'marginally toxic', where the mean *Corophium colo* survival was more than 80% of the control. McCready *et al.* (2005) found that the mean *Corophium colo* survival decreased slightly with increasing contamination and mean ERM quotients for sediments from Sydney Harbour. However, there was relatively low sensitivity in amphipod survival compared to sensitivity reported in previous United States data when sediment guidelines were exceeded. This may be a result of differences in sediment geochemistry.

**McCready et al. (2005)** suggested that metals may be the more important contaminant causing toxicity in the study. Results from the reburial tests showed more sensitivity, with a higher significant response observed in up to 70% of moderately contaminated sediments (39% toxic samples and 24% highly toxic samples to burial). An increase in responses in the reburial test was observed as contamination increased. In addition, a 96-hour water-only experiment with cadmium chloride was carried out as a positive control. *Corophium colo* was exposed to four to five unspecified concentrations of cadmium chloride and unspiked control. There were two replicates and ten individuals in each replicate. McCready *et al.* (2005) recorded a mean 96-hour LC50 value of 1.9 mg/l. Juveniles were found to be more sensitive than adults in the cadmium chloride experiment, with a recorded 96-hour LC50 of 0.6 mg/l for juveniles.

Overall, McCready *et al.* (2005) study suggests that although *Corophium colo* can effectively indicate acute toxicity in highly contaminated sediments, especially in reburial behaviour and when juvenile individuals are used, the species may be less sensitive than American amphipods in detecting lower levels of contamination.



**Montero et al. (2013a)** studied the effect of contaminated sediment from the Oiartzun estuary, in the Bay of Biscay on *Corophium multisetosum* through a laboratory-based whole sediment acute toxicity test. Sediment grab samples were collected from four stations (C, P, L, H) in the Oiartzun estuary along a contamination gradient and stations were classified based on the degree of toxicity; highly toxic, moderately toxic, and non-toxic. A sample was identified as toxic when there was a statistical difference of >20% mortality between control and contaminated sediment samples. The sediment was analysed for organic matter and trace metal (cadmium (Cd), copper (Cu), zinc (Zn) and lead (Pb) concentrations. The dissolved labile metal concentrations were also measured. *Corophium multisetosum* was exposed to the contaminated sediment samples in test beakers containing filtered natural seawater for 10 days. There were 20 individuals added to each test beaker. Mortality was recorded at the end of exposure.

Montero *et al.* (2013a) found a statistically significant difference between the stations for all metals found in the sediment, suggesting the sediment is toxic. Station C (located in the channel of the estuary) had the lowest concentration of heavy metals with 0.40 mg/kg of Cd, 86.15 mg.kg of Cu, 249.57 mg/kg of Zn and 30.43 mg/l of Pb in the sediment. The stations in the inner part of the estuary (P, L and H) had the highest metal concentrations. Station P (located in the middle part of the estuary) had metal concentrations of 0.69 mg/kg of Cd, 171.65 mg/kg of Cu, 473.32 mg/kg of Zn and 47.98 mg/kg of Pb in sediment samples . Station L sediments had metal concentrations of 0.60 mg/kg of Cu, 554.39 mg/kg of Zn and 62.15 mg/kg of Pb (Taken from Table 2). Station H sediments had metal concentrations of 1.34 mg/kg of Cd, 178.42 mg/kg of Cu, 750.32 mg/kg of Zn and 191.53 mg/kg of Pb . Therefore, the study classified station H as highly toxic, station P and L as moderately toxic and station L as non-toxic to *Corophium multisetosum*.

*Corophium multisetosum* mortality was significantly higher in the inner stations (P, L and H) compared to the control, with the highest mortalities observed at station H (around 90% mortality), followed by stations L and P (approximately between 50 to 60% mortality) (percentages from Figure 5, Montero *et al.*, 2013a). There was no significant difference in the mortality recorded at station C, the less contaminated station. Montero *et al.* (2013a) concluded that sediment contamination had a significant toxic effect on *Corophium multisetosum*, shown in the correlation between sediment metal concentrations and amphipod mortality.



**Montero** *et al.* (2013b) investigated the effects of contaminated sediment from three Spanish Atlantic harbours on *Corophium* sp. Sediment grab samples were collected from six sampling stations along a contamination gradient in Vigo (VI1, VI2, VI3, VI4, VI5), Bilbao (BI1, BI2, BI3, BI4, BI5) and Pasajes (PA1, PA2, PA3, PA4, PA5) harbours. Under laboratory conditions, *Corophium* sp. was exposed to sampled sediment in a 10-day whole sediment survival test. A sample was identified as toxic when there was a statistical difference of >20% mortality between control and contaminated sediment samples. Chemical analysis revealed sediments were contaminated with varying concentrations of heavy metals, PAHs, and PCBs (full list of concentrations in Table 1; Montero *et al.* (2013b). The concentrations of metals and organic compounds followed a gradient, with the highest contamination in the inner stations.

In Bilbao, station BI2 had the highest concentrations of all metals, except for Cd, where BI3 (1.07 mg/kg), BI4 (3.01 mg/kg) and BI5 (1.04 mg/kg) recorded slightly higher concentrations. The results found sediment samples from Bilbao had no significant toxic effect on Corophium sp. In Vigro, station VI2 had the highest contamination levels for all compounds, except station VI1, which had a higher concentration of Cu (751 mg/kg). Results found station VI2 had a significantly toxic effects on *Corophium* sp., with mortality approximately between 80 and 90% (Taken from Figure 2; Montero et al. (2013b). Station VI5 had mortality more than 20% higher than the control, but this increase was not statistically significant. In Pasajes, station PA2 was generally the most contaminated with high concentrations of Cu, Hg, Zn, PAHs, and PCBs. The results found sediment samples from stations PA2 had a significant toxic effect on *Corophium* sp. and had the highest percentage of mortality across all stations from all three harbours, with approximately 95 to 100% mortality. Stations PA3 and PA5 were also significantly toxic to Corophium sp., with approximately 30 to 40% mortality in PA3 and approximately 50% mortality in PA5 (Taken from Figure 2; Montero et al., 2013b). The results revealed that amphipod mortality was significantly correlated with chemical concentrations of Zn, PAHs, and PCB. Overall, the evidence showed that stations with high contamination of metals and/or organic compounds generally had a more significantly toxic effect, reducing the survival of Corophium sp.

**Nipper et al. (1998)** studied the effects of contaminated sediment from intertidal mudflats around the North Island of New Zealand on benthic communities and the chronic survival and growth of *Chaetocorophium lucasi*, using laboratory-based whole sediment toxicity tests. Sediment core samples were collected from four relatively uncontaminated sites: Aotea Harbour (site 1), Raglan Harbour (site 2), Manukau Harbour (site 3), Okura Estuary (site 4) and four contaminated sites; Tamaki Estuary (sites 5, 6 and7) and Manukau Harbour (site 8).



Sediment samples were analysed for heavy metals (cadmium, copper, lead, zinc, nickel, manganese, and iron), ammonium, pesticides (lindane, dieldrin, chlordane), polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs) and dichlorodiphenyl trichloroethanes (DDTs).

Laboratory-cultured juvenile *Chaetocorophium lucasi* was exposed to the contaminated sediment for 28 days in test jars with overlaying water that was replaced once a week. There were five replicates for each sediment sample and 14 individual *Chaetocorophium lucasi* in each replicate. Mortality was recorded after the exposure duration, and live individuals were retained for growth tests. Nipper *et al.* (1998) found the concentrations of heavy metals and organic chemicals were higher in sites 5 to 8. Site 8 had the highest metal concentration, and sites 5 and 7 had the highest organic chemical concentrations, particularly PAHs (see Tables 1 and 2; Nipper *et al.* (1998). *Chaetocorophium lucasi* survival significantly decreased in all sites, except site 8. Nipper *et al.* (1998) suggested that growth was not a useful endpoint for the study as there were no significant effects found in *Chaetocorophium lucasi* growth between sites.

A benthic community analysis was conducted during the study, using sediment cores from each site to assess changes in community structure across locations. *Chaetocorophium lucasi* was one of the most dominating marine organisms in the sediment samples. The amphipod was found abundant in control sediment samples, uncontaminated site 4 samples and contaminated sites 6 and 7 samples. This result was unexpected as samples from sites 6 and 7 caused high mortalities when tested in the laboratory. Nipper *et al.* (1998) concluded that none of the toxic tests responded more strongly to contaminated sediments than the reference uncontaminated sites, therefore neither natural sediment characteristics nor unmeasured contaminants affected the test organisms. It was noted that sediment collection and handling may have influenced results.

**Strode et al. (2017)** examined the effect of sediment toxicity on amphipod species *Bathyporeia pilosa* and *Corophium volutator* in the Gulf of Riga, Baltic Sea, using acute survival tests at 25 study sites between 2010 and 2012. The study assessed the toxic resistance of these species by exposing them to contaminated sediments under controlled laboratory conditions for 10 days, measuring survival rates as an endpoint. Bathyporeia pilosa exhibited survival rates ranging from 38% to 100%, while *Corophium volutator* had a survival range of 70 to 95%, indicating varied sensitivity between species. The findings suggested that sediment quality was mostly classified as low toxicity (64% of sites), though



some areas, particularly deeper central sites, showed moderate to poor quality. Strode *et al.* (2017) concluded that sediment contamination had minimal but detectable effects on amphipod populations and recommended integrating embryo malformation tests with acute toxicity assessments to improve monitoring. However, differences in species sensitivity may have influenced the survival rates, therefore further investigation would be needed into the environmental factors affecting amphipod responses to sediment toxicity.

**Van den Hurk et al. (1992)** examined the effect of contaminated North Sea sediments on infaunal amphipods, specifically *Bathyporeia sarsi* and *Corophium volutator*, using a 10-day static bioassay at multiple sites along two pollution gradients: one near an abandoned drilling site and another offshore of the Elbe-Weser plume in the German Bight. The amphipods were exposed to sediment samples with varying contamination levels, with mortality and sublethal effects (e.g., reburial ability, immobilization) recorded as endpoints. *Corophium volutator* exhibited significant mortality in sediments from the innermost stations of both gradients, particularly in the drilling site sediment (below 50% survival at Station A) and in multiple stations from the German Bight transect. *Bathyporeia sarsi* showed lower mortality rates of 12% but exhibited impaired reburial behaviour, with 28% of surviving individuals from Station A unable to rebury within one hour. The study concluded that the most contaminated sediments caused clear negative effects, though responses varied by species. The absence of a clear pollution gradient in toxicity data and inconsistencies in chemical analyses suggest potential confounding factors, such as sediment grain size and volatile contaminants.

**Van den Hurk et al. (1997)** examined the effect of dredged harbour canal sediments from the Rotterdam port area on the infaunal amphipod *Bathyporeia sarsi* through a 10-day whole sediment static bioassay at multiple sites, including an offshore disposal site in the North Sea. The study exposed *Bathyporeia sarsi* to sediments sampled from four locations in the Caland Canal (harbour area) and compared them to samples from an offshore disposal site ("North") and a cleaner reference site ("West"). Sediments from three of the four harbour sites (I, II, IV) resulted in significant mortality of *Bathyporeia sarsi*, with significant mortality figures ranging 28 to 100% mortality. However, although the offshore disposal site (site C) still exhibited toxicity in one sample, it was at a lower level than the harbour sites. Van den Hurk *et al.* (1997) concluded that sediments could still be toxic to benthic organisms despite them meeting Dutch sediment quality criteria and advised incorporating bioassays in dredge material assessments. They noted that toxicity at the disposal site was reduced compared to the harbour sites, likely due to dispersal of fine, contaminated sediments by strong currents,



transported to areas in the Wadden Sea. However, some residual toxicity remained, raising concerns about the environmental impact of offshore disposal.

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# 8 Sensitivity assessment

Sensitivity assessment uses the MarESA approach outlined in Tyler-Walters *et al.* (2023). The resistance, resilience and sensitivity terms and scales used here are defined in Tyler-Walters *et al.* (2024), and its application to contaminants evidence is outlined in Tyler-Walters *et al.* (2022).

#### 8.1 Recovery and resilience assessment

Corophium volutator is a mud shrimp with a long slender body up to 11 mm in length. The amphipod occupies semi-permanent U-shaped burrows up to 5 cm deep (Meadows & Reid, 1966) in the fine sediments of mudflats, saltmarsh pools and brackish ditches. It lives for a maximum of one year (Hughes, 1988) and females can have 2-4 broods in a lifetime (Conradi & Depledge, 1999). Populations in southerly areas such as the Dovey Estuary, Wales or Starrs Point, Nova Scotia have two reproductive episodes per year. Those populations in colder, more northerly areas such as the Ythan Estuary, Scotland or the Baltic Sea only have one (Wilson & Parker, 1996). On the west coast of Wales, breeding takes places from April to October and mating takes place in the burrow. Adult males crawl over the surface of the moist sediment as the tide recedes in search of burrows occupied by mature females. Corophium volutator forms an important food source for several species of birds and mobile predators such as fish and crabs (Hughes, 1988; Jensen & Kristensen, 1990; Raffaelli et al., 1991; Flach & De Bruin, 1994; Brown et al., 1999), so this behaviour makes them vulnerable to predation (Fish & Mills, 1979; Hughes, 1988; Forbes et al., 1996). The females can produce 20-52 embryos in each reproductive episode (Fish & Mills 1979; Jensen & Kristensen, 1990). Juveniles are released from the brood chamber after about 14 days, and development is synchronized with spring tides, possibly to aid dispersal. Recruitment occurs within a few centimetres of the parent, although they may disperse later by swimming (Hughes, 1988). In the warmer regions where Corophium volutator is found, juveniles can mature in two months (Fish & Mills, 1979) and add their own broods to the population. The juveniles born in May undergo rapid growth and maturation to reproduce from July to September and generate the next overwintering population (Fish & Mills, 1979).

*Corophium volutator* has great potential for recovery as it changes density and local distribution on an annual basis (Essink *et al.*, 1989; Flach & de Bruin, 1993; Hughes, 1988; Hughes & Gerdol, 1997; McLusky, 1968; Raffaelli *et al.*, 1991). Where perturbation causes local extinction (in areas on the scale of tens of square metres) *Corophium volutator* can



rapidly recolonize by immigration and recruitment of juveniles from immigrants. However, in areas of suitable habitat that are isolated from immigration, mass mortalities may have more serious implications for the recoverability of *Corophium volutator*. In the Ythan Estuary, where eutrophication led to the formation of beds of the gutweed *Ulva intestinalis, Corophium volutator* was almost completely eliminated from beneath it. However, high densities of *Corophium volutator* reappeared within a few months once the gutweed had disappeared in winter (Raffaelli *et al.*, 1991).

The amphipod genus Ampelisca has life history traits that allow them to recovery quickly where populations are disturbed. They do not produce large numbers of offspring but reproduce regularly and the larvae are brooded, giving them a higher chance of survival within a suitable habitat than free-living larvae. Ampelisca has a short lifespan and reaches sexual maturity in a matter of months allowing a population to recover abundance and biomass in a very short period (MES, 2008). Experimental studies have shown Ampelisca abdita to be an early colonizer, in large abundances of defaunated sediments where local populations exist to support recovery (McCall, 1977). Ampelisca abdita have been shown to migrate to, or from, areas to avoid unfavourable conditions (Nichols & Thompson, 1985). Mills (1967) reported that Ampelisca vadorum and Ampelisca abdita produced only one brood per generation but that there were two or more generations per year. In the English Channel, two reproductive patterns were identified. Species such as Ampelisca tenuicornis and Ampelisca typica produced two generations per year. The juveniles born in May-June were able to brood in September-October (Dauvin, 1988b; Dauvin, 1988c). Species such as Ampelisca armoricana and Ampelisca sarsi produced only one brood per generation and per year (Dauvin, 1989; Dauvin, 1988d). Ampelisca brevicornis showed an intermediate cycle with one generation per year during cold years (cold spring) and two generations per year during warm years (warm spring) and its cycle is intermediate between univoltine cycle and bivoltine cycle (Dauvin, 1988b,c,d,e; Dauvin, 1989; Dauvin & Bellan-Santini, 1990).

*Bathyporeia* spp. are short lived, reaching sexual maturity within 6 months with 6-15 eggs per brood, depending on species. Reproduction may be continuous (Speybroeck *et al.*, 2008) with one set of embryos developing in the brood pouch whilst the next set of eggs is developing in the ovaries. However, specific reproductive periods vary between species and between locations (Mettam, 1989) and bivoltine patterns (twice yearly peaks in reproduction) have been observed (Mettam, 1989; Speybroeck *et al.*, 2008). Adult amphipods are highly mobile in the water column and recolonization by the adults is likely to be a significant recovery pathway. The life history traits of rapid sexual maturation and production of multiple



broods annually support rapid local recolonization of disturbed sediments where some of the adult population remains.

Ampelisca spp. are very intolerant of oil contamination (Cabioch et al., 1978; Dauvin, 1987, 1998: Poggiale & Dauvin, 2001). The recovery of then Ampelisca populations in the fine sand community in the Bay of Morlaix took up to 15 years following the Amoco Cadiz oil spill in 1978 (Poggiale & Dauvin, 2001). Recolonization was delayed by the species' demography, low fecundity, lack of pelagic larvae, habitat isolation and the absence of local unperturbed source populations. Ampelisca armoricana began recolonization in 1981 but remained at low density until 1987, while Ampelisca sarsi actively recolonized in 1987, and Ampelisca tenuicornis (the most affected species) did not return until 1988. By 1993, Ampelisca densities had reached pre-spill levels (>40,000 ind. m<sup>2</sup>). Poggiale & Dauvin concluded that pollution levels initially dictated recolonization success, followed by competition and environmental conditions. Dauvin (1987) reported similar finding of Ampeliscids after the Amoco Cadiz oil spill and noted that Corophium crassicorne and Bathyporeia elegans suffered severe declines. However, Dauvin (1987) also noted that Corophium crassicorne had not recovered by the end of their study in 1986, eight years after the spill, while Bathyporeia elegans recovered within three to five years after the spill due to their due to its greater mobility.

**Resilience assessment.** The selected amphipod species (*Corophium* sp., *Ampelisca* sp., and *Bathyporeia* sp.) are known to have long reproductive seasons during their short lifespans. Where perturbation removes a portion of the population or even causes local extinction (resistance 'None', 'Low' or 'Medium') resilience is likely to be '**High'** for as long as recruitment from neighbouring areas and/or adult migration is possible. However, in areas of suitable habitat that are isolated, where total extinction of the population occurs (resistance 'None') recovery is likely to depend on favourable hydrodynamic conditions that will allow recruitment from farther away and recruitment to re-colonize impacted area may take longer. However, once an area has been recolonized, restoration of the biomass of both characterizing species is likely to occur quickly and resilience is likely to be '**Medium'** (full recovery within 2-10 years). However, the effects of oil spills may be prolonged, especially where contamination remains in the sediment and/or where the spill affects a wide area and no local unaffected populations remain. Hence, recovery from oil spills may be delayed, and resilience is assessed as '**Low'** (recovery within 10 to 25 years).



### 8.2 Hydrocarbons and PAHs - Sensitivity assessment

The count of ranked worst-case mortalities due to 'Hydrocarbons and PAHs' are summarized in Figure 3.1 and Table 8.1 below. The data presented in Table 8.1 include all life stages and articles where life stage were not reported.

Hydrocarbons and PAHs were reported to cause mortality in 78% of the results, of which 36% were 'severe' mortality and 38% was 'significant' based on the 36 articles examined. However, sublethal was reported in 16% of the results.

#### 8.2.1 Oil spills

Numerous studies reported the sensitivity of amphipods to oil spills (Suchanek, 1993). In particular, Ampelisca spp., Bathyporeia sp. and Corophium sp. were reported to be highly susceptible to oil pollution due to their sediment-dwelling habit and low dispersal (Cabioch et al., 1978; Dauvin, 1987, 1998; Gesteira & Dauvin, 2000; Poggiale & Dauvin, 2001). Amphipods were also reported to suffer declines after other oil spills. For example, significant declines in Corophium sp. after the Exxon Valdez oil spill (Wolfe, 1996) and Rose Bay oil spill (Roddie et al., 1994); and a 'sharp decline' in Ampelisca sp. after the Sea Empress spill (Nikitik & Robinson, 2003). Rostron (1998) also concluded that amphipods were sensitive to the effects of the Sea Empress spill. Smith (1968) reported that Corophium declined in heavily contaminated sites after the *Torrey Canyon* spill due to oil and dispersants. Widbom & Oviatt (1994) also reported significant declines in Ampelisca after the World Prodigy oil spill but no significant effects on Corophium. Kingston et al. (1997) reported that amphipods were completely absent from heavily impacted sites after the *Braer* oil spill and all sites in the path of the oil had reduced numbers. Dauvin (1987) reported that Bathyporeia experienced severe declines after the Amoco Cadiz oil spills. However, Dauvin (1987) also reported that Bathyporeia recovered within three to five years due to its high mobility, while Corophium crassicorne had not returned after ca eight years, and Poggiale & Dauvin (2001) reported that recovery in Ampelisca sp. took up to 15 years.

Overall, 76% of the results reported 'severe' mortality, and 11.5% reported 'significant' mortality due to oil spills across the selected amphipods examined. The weight of evidence suggests that Amphipoda are highly sensitive to oil spills. However, recovery rates may vary between species but depend on the time taken for the contamination to disperse (or be removed) and recruitment from unaffected populations.


Table 8.1. Summary of count of worst-case ranked mortalities to 'Hydrocarbons and PAH' contaminants reported in the evidence review and resultant proposed sensitivity assessments for selected amphipods (N= None, VL= Very low, L= Low, M= Medium, H = High, and NS= Not sensitive).

Group/Type/Genus	Severe	Significant	Some	None (rpt.)	Sublethal	Total	Resistance	Resilience	Sensitivity
Hydrocarbons (Petrochemical)									
Oil spill	22	3		1		26	Ν	L	н
Ampelisca	17	2				19	Ν	L	Н
Bathyporeia	2					2	Ν	М	Μ
Corophium	1	1		1		3	L	L	Н
Monocorophium	2					2	Ν	L	Н
Aromatics	1				1	2	Ν	Μ	Μ
Ampelisca	1					1	Ν	Μ	Μ
Corophium					1	1	Н	Н	NS
Complex	4	7	2	3	4	20	Ν	М	Μ
Corophium	4	7	2	3	4	20	Ν	М	Μ
Mixture	1				2	3	Ν	Μ	Μ
Corophium	1				2	3	Ν	Μ	Μ
Phenols	1	3			4	8	Ν	Μ	Μ
Ampelisca	1					1	Ν	М	Μ
Corophium		3			4	7	L	М	Μ
Total	12	13	2	4	28	59	Ν	Μ	Μ
Dispersant									
Dispersant	2		1	1		4	Ν	Μ	Μ
Corophium	2		1	1		4	Ν	Μ	Μ
Total	2		1	1		4	Ν	Μ	Μ
Hydrocarbons (pyrogenic)									
Polyaromatic hydrocarbons		20			3	23	L	Μ	Μ
Ampelisca		2			3	5	L	Μ	Μ
Corophium		18				18	L	Μ	Μ
Total		20			3	23	L	Μ	Μ
Grand Total	14	33	3	5	31	86	Ν	М	М

Therefore, the **resistance of amphipods to oil spills is assessed as 'None'**. The direct evidence of recovery after the *Amoco Cadiz* oil spills suggests that resilience should be



assessed as **'Medium'** in *Bathyporeia* but **'Low'** for *Ampelisca* and *Corophium*. Therefore, **sensitivity to oil spills** is assessed as **'Medium'** for *Bathyporeia* but **'High'** for *Corophium* and *Ampelisca*.

#### 8.2.2 Petroleum hydrocarbons

Fourteen of the articles examined the effects of petroleum oils (e.g. crude oil and fuel/bunker oils), and dispersed oils. Experimental studies examined the effects of oils, WAF or DWAFs in water-only or spiked sediment assays, in most cases using *Corophium* as the test organism, and only one article examined *Ampelisca*.

The reported toxicity varied between studies, types, concentration, and form of oil fractions tested. However, 20% of the results reported 'severe' mortality, 35% reported 'significant' mortality, and 10% reported 'some' mortality, while 'no' mortality was reported in 15% of the results. For example, the abundance of *Corophium* was significantly lower in the oiled treatments compared to controls, and experienced 'severe' mortality in sediment spiked with 60 mg/kg of Maya crude oil (Sanz-Lázaro & Marín, 2009).

McClusky (1982) reported that *Corophium volutator* was absent within 500 m of the petrochemical effluent discharge in the intertidal mudflats of the Kinneil area, Forth Estuary, Scotland. McClusky & Martins (1998) reported that *Corophium* populations on the Kinneil mudflats increased in 1985 after the closure of the phenol plant. They noted that *Corophium* abundance was correlated significantly with a decrease in chemical effluents from the phenol or acrylonitrile plants and the resultant decrease in BOD (biological oxygen demand).

Van Eenennaam *et al.* (2018) simulated the effects of the Marine Oil Snow Sedimentation and Flocculent Accumulation (MOSSFA) event that occurred after the *Deepwater Horizon* (DWH) oil spill. The MOSSFA transported ca 14% of the total released oil to the seabed. The oil treatment (10 g/m<sup>2</sup> for 16 days in laboratory mesocosms) severely reduced survival to 9% of controls (91% mortality) while the 'snow and oil' treatment reduced survival to 20% (80% mortality), probably because *Corophium* escaped the snow layer on the sediment and possibly fed on the artificial snow.

Oil fractions also modified behaviour. Percy (1977) reported that *Corophium clarencense* actively avoided oil-spiked sediments in choice experiments. Kienle & Gerhardt (2008) noted that WAF affected locomotor activity and burial in *Corophium volutator*. Scarlett *et al.* (2007) reported that reproduction was significantly less in the DWAF and all oil treatments than in controls, Corexit and WAF treatments.

In his review, Suchanek (1993) identified amphipods, particularly ampeliscid species, as highly sensitive to oil contamination, often experiencing severe population declines and failing to recover for at least five years post-spill, likely due to the longevity of oil in sediments. Therefore, **the worst-case resistance to petrochemical hydrocarbons is assessed as 'None' for amphipods as a group**, and in particular *Corophium*. No evidence on the resistance of *Bathyporeia* was found. Hence, resilience is assessed as **'Medium'** and the **worst-case sensitivity to petrochemical hydrocarbons in the selected amphipods** is assessed as **'Medium'**.

#### 8.2.3 Dispersants

Scarlett *et al.* (2005, 2007) studied the toxicity of dispersants. Scarlett *et al.* (2005) examined the effects of the dispersants Corexit 9527 and Superdispersant-25 (SD-25) on a selection of invertebrates, including *Corophium volutator*. Mortality was 100% at 500 ppm of both dispersants after 48 hours, and at 375 ppm in Corexit 9527. The mortality from exposure to Corexit 9527 was significantly greater than SD-25, and recovery rates of survivors from SD-25 were higher than from Corexit 9527. Scarlett *et al.* (2005) noted that dispersant concentrations of ≥175 ppm reduced burrowing in the laboratory, which, in the wild, might allow *Corophium* to leave contaminated sediments. Scarlett *et al.* (2007) reported some (25%) mortality in adults exposed to Corexit 9527 in acute tests but no significant mortality in neonates in chronic tests.

Overall, the evidence suggests that *Corophium* is sensitive to the dispersants examined, although no information on *Bathyporeia* or *Ampelisca* was found. Therefore, **the worst-case resistance of selected amphipods to dispersants** (Corexit 9527 and Superdispersant-25) is **assessed as 'None'**. Resistance is probably **'Medium'**, so the **worst-case sensitivity to dispersants is assessed as 'Medium'** but with 'Low' confidence due to the limited number of studies found.

## 8.2.4 Polyaromatic hydrocarbons (PAHs)

The articles reviewed examined the toxicity of 14 different PAHs or total PAH resulting from contamination. PAH exposure was reported to result in 'significant' mortality in 87% of worst-case results examined, and only sublethal effects in 13% of the results examined (Table 8.1). The majority of the results were derived from 10-day LC50s in sediment bioassays.

For example, Boese *et al.* (1997) concluded that photoactivation significantly increased the toxicity of fluoranthene. In *Corophium insidiosum*, fluoranthene exposure alone resulted in a



96-hour LC50 of 85  $\mu$ g/l, which decreased to 32  $\mu$ g/l after UV exposure, while the EC50 for reburial declined from 54  $\mu$ g/L to 20  $\mu$ g/l, indicating an increase in toxicity after UV exposure. Similarly, Spehar *et al.* (1999) reported that fluoranthene was toxic under fluorescent light, with a 96-hour LC50 of 67  $\mu$ g/l (59 to 76  $\mu$ g/l confidence interval) in *Ampelisca abdita*, but under UV exposure, toxicity increased significantly, leading to complete mortality at concentrations that were too low to determine an LC50.

In sediment bioassays, Fisher *et al.* (2011) determined acute toxicity values (10-day LC50) for 12 PAHs in *Corophium volutator*, which ranged from 24 mg/kg for 4methyldibenzothiophene to over 1,000 mg/kg for anthracene (see evidence summary spreadsheet). Fisher *et al.* (2011) concluded that PAH contamination in sediments can pose a significant risk to benthic organisms, with certain compounds demonstrating much higher toxicity than others. Swartz *et al.* (1990) exposed *Corophium spinicorne* and *Rhepoxynius abronius* to fluoranthene in sediment. *Corophium spinicorne* was less sensitive to fluoranthene exposure than *Rhepoxynius abronius*, with an interstitial water 10-day LC50 of 37.9 µg/l compared to 23.8 µg/l for *Rhepoxynius abronius*. Swartz *et al.* (1990) concluded that *Corophium spinicorne* was partly protected from the contaminated interstitial water because it lives in burrows connected to the overlying water. Ciarelli *et al.* (1999) noted that PAH exposure affected burrowing activity, and hence bioturbation, in *Corophium volutator*.

Overall, the **worst-case resistance to PAH exposure** in *Corophium* sp. and *Ampelisca* sp. is **assessed as 'Low'** based on the evidence above. Resilience is probably '**Medium'**, so **sensitivity is assessed as 'Medium'**.

## 8.2.5 Petroleum hydrocarbons – others

Brown *et al.* (1999) reported that exposure to high concentrations of 5-nonylphenol resulted in mortality while lower concentrations resulted in significant reduction in both survival and growth and interfered with reproduction, with potential population effects. Redmond & Scott (1987) reported 100% ('severe') mortality in *Ampelisca* after exposure to 150 mg/l of phenol. In mating experiments, Krang (2007) reported that exposure to 0.5 and 5  $\mu$ g/g naphthalene significantly reduced the ability of males to locate mature females, which could prevent mating and reproduction. The population dynamics of amphipods are marked by rapid population growth and significant predation. A reduction in mating success could significantly affect amphipod populations and the populations of other species dependent on them for food.



Overall, the limited evidence above suggests that *Corophium* and *Ampelisca* have **'Low'** and **'None' resistance** to nonylphenol and phenol, respectively. Hence, resilience is assessed as 'Medium' and **sensitivity as 'Medium'** but with 'Low' confidence.

# 8.3 Transitional metals and organometals – sensitivity assessment

The count of ranked worst-case mortalities due to 'Transitional metals and organometals' are summarized in Figure 4.1 and Table 8.2 below. The data presented in Table 8.2 include all life stages and articles where life stage were not reported.

#### 8.3.1 Transitional metals

Transitional metal exposure was reported to result in 'severe' mortality in 11.4% of worstcase results examined, 'significant' in 73.4%, 'some' in 5.7%, 'no' mortality in 3.8% and only sublethal effects in 5.7% of the results examined (Table 8.2). The majority of studies used amphipods as a standard bioassay, based on standard 96-hour or 10-day assays, to derive LC50 endpoints. Hence, the results are dominated by 'significant' (i.e. 50%) mortality endpoints.

Metal toxicity was shown to vary with salinity, temperature, pH, and season, as well as between species (Bigongiari *et al.*, 2004; Kater *et al.*, 2000; Prato *et al.*, 2008; Re *et al.*, 2009; Roberts *et al.*, 2013). Roberts *et al.* (2013) concluded that exposure to pCO<sub>2</sub> (ocean acidification) increased the toxicity of the metal contaminated sediment, probably due to the effects of pH on their physiology rather than metal speciation. Shipp & Grant (2006) suggested that *Corophium* was more exposed to Cu in pore water than in sediment because of it lives in U-shaped burrows. Pérez-Landa *et al.* (2008) reported that *Corophium multisetosum* was more sensitive to ammonia and Cd in summer and less sensitive in the winter.

Strode & Balode (2013) was the only article to examine the effects of metal exposure on *Bathyporeia* directly. Sensitivity to metal exposure (Cd, Cu and Zn) varied significantly between species, while Cd was the most toxic metal tested. They reported that *Bathyporeia pilosa* exhibited moderate resistance compared to other tested amphipods, with the freshwater species *Gammarus pulex* being among the most sensitive.



Table 8.2. Summary of count of worst-case ranked mortalities to 'Transitional metals and organometals' contaminant reported in the evidence review and resultant proposed sensitivity assessments for selected amphipods (N= None, VL= Very low, L= Low, M= Medium, H = High, and NS= Not sensitive).

Group/Type/Genus Metals	Severe	Significant	Some	None (rept.)	Sublethal	Total	Resistance	Resilience	Sensitivity
Metals & their compounds	18	116	9	6	9	158	Ν	Μ	М
Ampelisca	3	25			1	29	Ν	М	М
Bathyporeia		6				6	L	М	М
Chaetocorophium	2	2				4	Ν	М	М
Corophium	13	80	9	6	8	116	Ν	М	М
Gammarus		2				2	L	М	М
Paracorophium		1				1	L	М	М
Mixture		2	1			3	L	М	Μ
Bathyporeia		1				1	L	М	М
Corophium		1	1			2	L	М	М
Nanoparticulates	1	3	1	3	2	10	Ν	М	Μ
Corophium	1	3	1	3	2	10	Ν	М	М
Total	19	121	11	9	11	171	Ν	Μ	Μ
Organometals									
Organotin		2				2	L	М	Μ
Corophium		2				2	L	М	М
Grand Total	19	123	11	9	11	173	Ν	М	М

Strode *et al.* (2017) examined the effect of metal contaminated sediment on *Bathyporeia pilosa* and *Corophium volutator. Bathyporeia pilosa* exhibited survival rates ranging from 38% to 100%, while *Corophium volutator* had a survival range of 70 to 95%, indicating varied sensitivity between species, although the sediment was described as low toxicity. Warwick (2001) noted that *Corophium volutator* was absent from the heavy metal polluted areas of the Fal estuary, Cornwall.

Overall, 'severe' mortality was reported in 11.4% of the results, including *Corophium* and *Ampelisca* and 'significant' mortality in 73.4% of the results examined, including *Bathyporeia* (Table 8.2). Therefore, the **worst-case resistance to transitional metal exposure is** 

assessed as 'Low' in *Bathyporeia* and 'None' in *Corophium* and *Ampelisca*. Resilience is probably 'Medium' for all species, and worst-case sensitivity to transitional metal exposure is assessed as 'Medium' in *Bathyporeia*, *Corophium* and *Ampelisca*.

#### 8.3.2 Organometals

Stronkhorst *et al.* (1999) was the only study of the effects of TBT on *Corophium volutator* found. They reported 10-day LC50s at low (ng) levels of TBT in both pore water (329 ng/l TBT) and spiked sediment (2,185 ng/g TBT). They concluded that TBT was very toxic to *Corophium volutator,* especially in pore water. No evidence on *Ampelisca* or *Bathyporeia* was found. Therefore, **the worst-case resistance to organotin is assessed as 'Low'**, resilience as 'Medium' **and sensitivity as 'Medium**. The assessment is probably applicable to all three species of amphipod based on similarities in their physiology and endocrinology, but confidence in the assessment is 'Low' as evidence is limited to a single article.

#### 8.3.3 Nanoparticulate metals.

The toxicity of zinc oxide nanoparticulates (ZnO NPs) was studied in only four papers. In all cases, the articles compared the toxicity of ZnO NPs with inorganic zinc, as ZnO, ZnCl<sub>2</sub> or ZnSO<sub>4</sub> (Fabrega *et al.*, 2012; Larner *et al.*, 2012; Vimercati *et al.*, 2020; Righi *et al.*, 2023). In all cases, the authors concluded that zinc nanoparticulates were no more toxic than inorganic zinc and that the toxicity was due to Zn ions and that dissolution of the nanoparticulates contributed to their toxicity. The articles reported a range of mortalities from 'severe' to 'none', depending on the concentration tested. For example, Vimercati *et al.* (2020) reported a 96-hour LC50 for ZnO NPs of 1.75 mg/l, and Righi *et al.* (2023) reported a 96-hour LC50 of 1.93 mg/l ZnO-NP in *Corophium insidiosum.* However, Larner *et al.* (2012) reported >80% survival in *Corophium volutator* exposed to ZnO-NPs (around 1,000 ng/g Zn). Nevertheless, Fabrega *et al.* (2012) noted that all forms of zinc tested significantly affected growth and could have long-term effects on populations of *Corophium volutator*.

Righi *et al.* (2023) also examined the toxicity of calcium phosphate nanoparticles (CaP-N). They concluded that CaP-N) were not toxic to *Corophium insidiosum* and *Gammarus aequicauda* as their 96-hour LC50s were >100 mg/l. They also noted that *Corophium insidiosum* incorporated the CaP-N nanoparticles into their tubes during the 96-hour experiment. However, no evidence of the effects of other metal nanoparticulates was found.



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Overall, the **worst-case resistance to the effects of zinc nanoparticulates** is assessed as **'None'**. Resilience is probably **'Medium'**. Hence, the **worst-case sensitivity** is assessed as **'Medium'**. Confidence in the assessment is 'Low' due to the limited number of studies found.

# 8.4 Synthetic compounds – sensitivity assessment

The count of ranked worst-case mortalities due to 'Synthetic compounds' are summarized in Figure 5.1 and Table 8.3 below. The data presented in Table 8.3 include all life stages and articles where life stage were not reported.

#### 8.4.1 Pesticides/biocides

Pesticide exposure was reported to result in 'severe' mortality in 11.5% of worst-case results examined, 'significant' in 60.6%, 'some' in 0%, 'no' mortality in 0% and only sublethal effects in 27.8% of the results examined (Table 8.3).

'Severe' (>75%) mortality was reported in five articles. Dumbauld *et al.* (2001) reported a 97% loss in abundance of *Corophium* in their large-scale experimental treatment of mudflats with carbamate pesticide, used to control mud and ghost shrimp, although not in all of their experiments. Collier & Pinn (1998) reported 100% mortality in *Corophium volutator* exposed to 80.0 mg/m<sup>2</sup> of ivermectin for 14 days (the highest concentration tested). Similarly, Davies *et al.* (1998) reported >92% mortality after exposure to sediment spiked with ≥0.7 mg/kg of ivermectin after 10 days, with a 10-day LC50 value of 0.18 mg/kg. Mayor *et al.* (2008) reported severe mortality in *Corophium volutator* due to exposure to several pesticides (cypermethrin, emamectin benzoate, and azamethipos). Thursby & Berry (1987; cited from ECOTOX) reported 100% mortality in *Corophium* after 96-hour exposure to 37.5 µg/l of the organophosphate pesticide hydroxydiazinon. 'Significant' mortality (25-75%) was reported by another 15 articles, mainly as LC50s based on 24-, 48-, 96-hour or 10-day bioassays.

Overall, 72% of the worst-case results examined reported 'severe' to 'significant' mortality due to pesticide exposure. Hence, the **worst-case resistance to pesticide exposure** is assessed as **'None'**. Resilience is probably **'Medium'**, so sensitivity is assessed as **'Medium'**.



Table 8.3. Summary of count of worst-case ranked mortalities to 'Synthetic compound' contaminants reported in the evidence review and resultant proposed sensitivity assessments for selected amphipods (N= None, VL= Very low, L= Low, M= Medium, H = High, and NS= Not sensitive).

	/ere	nificant	ne (rept.)	olethal	al	sistance	silience	nsitivity
Group/Type	Sev	Sig	Noi	Sul	Tot	Re	Re	Ser
Pesticide/Biocide								
Antifoulant		2		1	3	L	М	М
Antimicrobial				1	1	Н	Н	NS
Carbamate	1	1		9	11	Ν	Μ	М
Fungicide		1		1	2	L	Μ	М
Herbicide		11			11	L	Μ	М
Insecticide		1		1	2	L	Μ	М
Organohalogen		11		3	14	L	М	М
Organophosphate	2	3		1	6	Ν	М	М
Parasiticide	3	6			9	Ν	М	М
Pyrethroid	1	1			2	N	М	М
Pesticide/Biocide Total	7	37		17	61	N	Μ	Μ
Pharmaceutical								
Analgesic (NSAID)		1			1	L	Μ	М
Antibiotic	1				1	Ν	Μ	М
Antidepressant		1			1	L	М	М
Pharmaceutical Total	1	2			3	Ν	М	Μ
PCBs								
PCBs		5			5	L	М	М
Phthalates								
Phthalate esters	1	2		1	4	L	Μ	М
PPCPs								
Ultraviolet (UV) filter		2			2	L	М	М
Synthetics (other)								
Alcohols	1				1	N	М	М
Glycols			1		1	Н	Н	NS
Surfactant	1	3	1		5	N	М	М
Synthetics (other) Total	2	3	2		7	N	Μ	М
Total	10	51	2	18	82	Ν	Μ	Μ



#### 8.4.2 Pharmaceuticals

Only two articles examined the effects of pharmaceuticals on the amphipods reviewed. Mayor *et al.* (2008) reported 93% mortality in *Corophium* exposed to sediment spiked with 100 mg/kg (the highest concentration tested) of the antibiotic oxytetracycline in 10-day bioassay and a 10-day LC50 of 414  $\mu$ g/kg of oxytetracycline.

Gronlund *et al.* (2004) determined 10-day LC50s for the analgesic diclofenac and antidepressant citalopram in spiked sediment bioassays. They noted that *Corophium volutator* mortality increased drastically in a narrow range of concentrations. Diclofenac was more toxic than citalopram to *Corophium volutator*. However, Gronlund *et al.* (2024) suggested that citalopram is unlikely to directly pose a lethal threat to the *Corophium volutator* community based on citalopram concentrations in water and sediment/water partitioning observed in the experiments. Results from the mixture study found a synergistic effect, meaning the combined impact of the contaminants was greater than expected (Gronlund *et al.* 2024).

Overall, the results indicate that the **worst-case resistance to pharmaceutical exposure** should be assessed as **'None'**. Hence, resilience is probably 'Medium' and **sensitivity is assessed as 'Medium'**. However, the number of studies, species, and pharmaceuticals tested was limited, so the assessment is made with 'Low' confidence.

## 8.4.3 Polychlorinated biphenyls (PCBs)

Only two of articles reviewed examined the effects of PCBs on the selected amphipods. They reported significant mortality in the form of LC50s (Table 8.3). Reish (1993) reported a 96-hour LC50 of 0.009 mg/l PCB (Arochlor 1254) in water-only assays of *Corophium insidiosum*. Ho *et al.* (1997) reported that Aroclor 1242 was more toxic to *Ampelisca abdita* than Aroclor 1254. The 96-hour LC50 value for Aroclor 1242 was 10 ppb, and for Aroclor 1254 was 40 ppb, respectively. Ho *et al.* (1997) concluded that PCBs were likely to be the source of chronic and reproductive toxicity in the New Bedford Harbor (NBH) sediments studied.

Overall, 'significant' mortality due to PCB exposure was reported in both studies and both PCBs examined. Therefore, **the worst-case resistance to PCB exposure** is assessed as **'Low'**, resilience as **'Medium'** and **sensitivity as 'Medium'**. However, the number of studies, species, and PCBs tested was limited, so the assessment is made with 'Low' confidence.



## 8.4.4 Phthalates

Only three of the articles reviewed examined the effects of Phthalates (phthalate esters) on the selected amphipods. Tagatz *et al.* (1983) examined the effect of di-n-butyl phthalate (DBP) exposure on communities that colonized sediment in the laboratory. The number of *Corophium acherusicum* significantly decreased to eight individuals in the 0.34 mg/l DBP treatment. There were zero *Corophium acherusicum* individuals recorded in the 3.7 mg/l DBP treatment. *Corophium acherusicum* in laboratory colonized communities were significantly affected by as low a concentration as 0.34 mg/l DBP. Tagatz & Stanley (1987; cited from ECOTOX) reported a 96-hour LC50 of 450 µg/l DBP in *Corophium acherusicum*. Ho *et al.* (1997) examined the toxicity of bis(2-ethylhexyl) phthalate on *Ampelisca abdita*. They reported a 48-hour LC50 above 1,000 ppb of phthalate for *Ampelisca abdita* and suggested that the phthalate was not toxic to amphipods in the samples studied.

Therefore, **the worst-case resistance to phthalate exposure** is assessed as **'None'**, resilience as **'Medium'** and **sensitivity as 'Medium'**. However, the number of studies, species, and phthalates tested was limited, so the assessment is made with 'Low' confidence.

## 8.4.5 Synthetics (other)

Only six articles examined the toxicity of 'other' synthetic chemicals in the selected amphipods. Hester *et al.* (1991) examined the effects of Arosurf MSF (monomolecular surface film) on a range of marine species including *Gammarus* sp. No significant mortality was observed in any of the species tested, after 96 hours at 47 ml/m<sup>2</sup> of Arosurf MSF.

Picone *et al.* (2008) and Lera *et al.* (2008) examined the toxicity of sodium dodecyl sulphate (SDS) surfactants in *Corophium orientale* and reported 96-hour LC50s of 8.7 mg/l SDS and 3.14 mg/l SDS, respectively. Rico-Rico *et al.* (2009) examined the toxicity of the surfactant C12-2-LAS (2-(*p*-sulphophenyl)-dodecane) to *Corophium volutator.* They reported a 5-day LC50 of 0.73 mg/l but also noted that mortality was not significant at 50 mg/l but increased to 100% at 500 mg/l.

Therefore, **the worst-case resistance to surfactants may be considered 'None'** (Rico-Rico *et al.*,2009) or **'Low'** (Lera *et al.*, 2008; Picone *et al.*, 2008) depending on the surfactant. In both cases, resilience is probably **'Medium**', so **sensitivity is assessed as 'Medium'**. However, the number of studies, species, and chemicals tested was limited so the assessment is made with 'Low' confidence.





Thursby & Berry (1987) examined the toxicity of Triethylene glycol diacetate on *Ampelisca abdita* and reported no mortality after 96 hours at 0.5  $\mu$ g/l but did not specify mortality at 156  $\mu$ g/l. Harris & Morgan (1984) reported that 1% ethanol was lethal to *Corophium volutator* after 24 hours. Fastelli & Renzi (2019) examined the toxicity of chemical-based sunscreen and nanoparticulate metal physical-based (TiO2-NPs; ZnO-NPs) sunscreen on *Corophium orientalis*. The 96-hour LC50 was 82  $\mu$ l/l for chemical-based sunscreen and 87  $\mu$ l/l for physical-based sunscreen at 4.0 ppt salinity. Fastelli & Renzi (2019) concluded that chemical-based and physical-based sunscreen types can exert different ecotoxicological responses on the tested species, suggesting that sensitivity toward nanoparticles in personal care products could be very different amongst different marine taxa.

Worst-case resistance and sensitivity assessments for glycols and sunscreens are suggested in Table 8.3. However, the number of studies, species, and chemicals tested was limited, so the assessment is made with 'Low' confidence.

# 8.5 Other substances

The count of ranked worst-case mortalities due to 'Other substances' are summarized in Table 8.4 below. Most studies reported 'significant' mortality as LC50s and are listed in the evidence summary spreadsheet. Ammonia was the most studied chemical in relation to nutrients water quality and is outside the scope of this study.

Hydrogen peroxide was examined as a disinfectant in ballast water. Smit *et al.* (2008) concluded that *Corophium volutator* had a relatively low sensitivity to hydrogen peroxide compared to *Artemia salina, Brachionus plicatilis, Dunaliella teriolecta*, and *Skeletonema costatum*, which were the other species tested in the study. Caldwell (1975) concluded that *Corophium salmonis* are unable or barely able to tolerate 1.0 mg/l of hydrogen sulphide (as sodium sulphide) during continuous exposure for multiple days. Ferretti *et al.* (2004) examined the suitability of KCI as a reference toxicant in bioassays. Soto *et al.* (2000) exposed *Ampelisca araucana* juveniles to 38.5, 55, 78.5, 112, and 160 mg/l of potassium dichromate for 48 hours, resulting in a 48-hour LC50 value of 56.89 mg/l.



Table 8.4. Summary of count of worst-case ranked mortalities to 'Other substances' contaminants reported in the evidence review and resultant proposed sensitivity assessments for selected amphipods (N= None, VL= Very low, L= Low, M= Medium, H = High, and NS= Not sensitive).

Group/Type	Severe	Significant	Sublethal	Total	Resistance	Resilience	Sensitivity
Inorganic chemicals							
Ammonia	1	7		8	Ν	М	М
Hydrogen peroxide			2	2	Н	Н	NS
Potassium chloride		1		1	L	М	М
Potassium dichromate		1		1	L	М	М
Sodium sulphide		3		3	L	М	М
Total	1	12	2	15			

Worst-case resistance and sensitivity assessments for each chemical are suggested in Table 8.4. However, the number of studies, species, and chemicals tested was limited so the assessment is made with 'Low' confidence.



# 9 Conclusions

This report presents the finding of a time limited Rapid Evidence Assessment (REA) of the effects of contaminants on selected amphipods, in particular, *Corophium* sp., *Ampelisca* sp. and *Bathyporeia* sp.

- The initial literature review returned ca 3,475 articles from several different amphipod species. The first screening resulted in over 600 articles that could not be processed in then time available to the review.
- Therefore, the review was strictly limited to *Bathyporeia* and *Corophium*, but *Ampelisca* was retained under 'Hydrocarbons and PAHs'. Hence, the review was limited to ca 143 articles.
- Most of the articles examined *Corophium* but *Bathyporeia* was notably poorly studied, as only nine articles were found in total during this review of the effects of contaminants.
- Most articles examined (45%) studied the effects of transitional metals on the amphipods. However, this number is skewed because most of the bioassay studies used inorganic cadmium (usually CdCl<sub>2</sub>) as a reference toxicant in their experiments.
- Petrochemical hydrocarbons (inc. oil spills) (15.6%) and 'pesticides/biocides' (15.4%) were the next most studied contaminant group.
- Amphipods are used to examine the toxicity of contaminant sediment. In this review, 7.6% of the results were derived from contaminated sediment bioassays.
- Oil spills were amongst the most toxic event to the selected amphipods with 86% of the results from the relevant studies reporting 'severe' or 'significant' mortality. Overall, 68% of the results for 'Hydrocarbons (petrochemical)' (including oil spills, discharges, and phenols) reported 'severe' or 'significant' mortality.
- Polyaromatic hydrocarbons (PAHs) (of pyrogenic origin) were similarly toxic with 86% of the results from the relevant studies reporting 'severe' or 'significant' mortality.
- Metals and their compounds (including nanoparticulates) were the next most toxic with 79% of the results from the relevant studies reporting 'severe' or 'significant' mortality.



- Pesticides / biocides were the next most toxic with 72% of the results from the relevant studies reporting 'severe' or 'significant' mortality.
- The results from studies of 'pesticides/biocides', polyaromatic hydrocarbons (PAHs), transitional metals and nanoparticulates were dominated by LC50 values.
- Contaminated sediments studies reported 'severe' or 'significant' mortality in 76% of the results examined but could not be included in sensitivity assessments as the sediments studied contained a variety of different contaminants (e.g. metals, PAHs, PCB, and pesticides).
- Several of the less well studied contaminant groups (e.g. pharmaceuticals, PCBs, phthalates and 'others') were represented by a limited number of studies, so confidence in those assessments is 'Low'.

It should be noted that this timed review focused on the effects of contaminants on a limited number of species and does not represent Amphipoda as a group. The range of contaminants addressed was also limited by the focus of the review. Over twice as many articles on amphipods were identified in the initial literature review than could be covered here. An adequate review of the effects contaminants on Amphipoda would require further study.



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