Title	The freshwater isopod Asellus aquaticus as a model biomonitor of environmental pollution: A review
Author(s)	O'Callaghan, Irene; Harrison, Simon; Fitzpatrick, Dara; Sullivan, Timothy
Publication date	2019-06-28
Original citation	O'Callaghan, I., Harrison, S., Fitzpatrick, D. and Sullivan, T. (2019) 'The freshwater isopod Asellus aquaticus as a model biomonitor of environmental pollution: A review', Chemosphere, 235, pp. 498-509. doi: 10.1016/j.chemosphere.2019.06.217
Type of publication	Article (peer-reviewed)
Link to publisher's version	http://www.sciencedirect.com/science/article/pii/S0045653519314584 http://dx.doi.org/10.1016/j.chemosphere.2019.06.217 Access to the full text of the published version may require a subscription.
Rights	**© 2019 Elsevier Ltd. All rights reserved. This manuscript version is made available under the CC BY-NC-ND 4.0 license.**
Item downloaded from	http://hdl.handle.net/10468/10143

The freshwater isopod *Asellus aquaticus* as a model biomonitor of environmental pollution: a review.

I. O'Callaghan[a,b], S. S. C. Harrison[a], D. Fitzpatrick[b] and T. Sullivan[a*]

[a] *School of Biological, Earth & Environmental Sciences, University College Cork, Ireland*
[b] *School of Chemistry, University College Cork, Ireland*
** Corresponding author*

Abstract

Anthropogenic substances pollute freshwater systems worldwide, with serious, long-lasting effects to aquatic biota. Present methods of detecting elevated levels of trace metal pollutants are typically accurate but expensive, and therefore not suitable for applications requiring high spatial resolution. Additionally, these methods are not efficient solutions for the determination of long-term averages of pollution concentration. This is the rationale for the implementation of a biomonitoring programme as an alternative means of pollutant detection.

This review summarises recent literature concerning the past and potential uses of the benthic isopod *Asellus aquaticus* as a biomonitor for pollution in freshwater systems. Recent studies indicate that *A. aquaticus* is well suited for this purpose. However, the mechanisms by which it bioaccumulates toxins have yet to be fully understood.

In particular, the interactions between coexisting trace metal pollutants in the aquatic environment have only recently been considered, and it remains unclear how a biomonitoring programme should adapt to the effects of these interactions. It is evident that failing to account for these additional stressors will result in an ineffective biomonitoring programme; for this reason, a comprehensive understanding of the bioaccumulation mechanisms is required in order to reliably anticipate the effects of any interferences on the outcome.

Keywords:
Asellus aquaticus; biomonitor; freshwater; trace metal; pollution.

Highlights:
- *Asellus aquaticus* is known to bioaccumulate trace metals
- *A. aquaticus* has been suggested as a potential biomonitor for metal pollutants
- A biomonitoring programme could act as an early warning system for pollution events
- Metal uptake and sequestering mechanisms are still poorly understood
- Antagonistic effects of metals must be better understood for implementation to succeed

Capsule:
"The benthic isopod, *Asellus aquaticus*, may be an ideal biomonitor for freshwater pollution."

I. Introduction

Pollutants, such as trace metals and pesticides, can cause detrimental effects when they enter into the freshwater environment. Metal pollutants, in particular, may enter the hydrosphere via a number of (primarily anthropogenic) pathways (Javed, 2005; Klavinš et al., 2000; Salt et al., 1995; Stead-Dexter and Ward, 2004). These metals may be toxic, even in small concentrations, to aquatic life, partly due to the tendency of fauna to bioaccumulate trace amounts (Palma et al., 2015). The mobility, transport and fractionation of a metal species are defined by the chemistry of the molecule, as well as the physicochemical and biological characteristics of the river system (Sakan et al., 2009). All of these parameters determine the bioavailability of the metal.

The monitoring of metal concentrations in waterways traditionally requires offline chemical analysis methods which may be costly, time-consuming and may not fully reflect the time-averaged conditions of the water system (Markert et al., 1999). This has led to a search for alternative methods of monitoring. The implementation of a biomonitor is a reasonable solution.

Benthic dwellers have been suggested as an ideal biomonitor of river and lake pollution, as they are generally present in large numbers, and have been found to strongly accumulate trace metals (El Gawad, 2009). As they do not typically displace themselves over large distances, they are considered to be a fair representation of conditions in the area in which they are found (Adriaenssens et al., 2006). Benthic species are often predated upon, potentially filling the role of a keystone species, and contamination of these species may have far reaching consequences for ecosystem health (Fleeger et al., 2003).

The sensitivity and tolerance of a particular species must be considered, as different species have been found to react differently to the presence of pollutants (De Jonge et al., 2008; MacNeil et al., 2002). In particular, the isopod species, *Asellus aquaticus* (*Fig. 1*), has been identified as a potential biomonitor, due to its greater tolerance for metal pollution (Whitehurst, 1991) and its wide range, which has not been greatly affected by human impact (Verovnik et al., 2005).

A. aquaticus has been widely studied, and the life history and population distribution of the species is well documented (Adcock, 1979; Sket, 1994; Steel, 1961; Williams, 1962). The species is increasingly employed as a biomonitor of environmental contamination and has been used for more novel, and recently identified, environmental pollutants, ranging from polycyclic aromatic hydrocarbons (PAHs) and endocrine-disrupting compounds, to radionuclide contaminants and tungsten carbide (WC) nanoparticles (De Lange et al., 2006; Ekvall et al., 2018; Fuller et al., 2018; Weltje and Oehlmann, 2006).

In this review, we define the term "biomonitor" to be synonymous with the description presented in a study by Rainbow in 1995, as "a species which accumulates heavy metals in its tissues, and may therefore be analysed as a measure of the bioavailability of the metals in the ambient habitat" (Rainbow, 1995).

The majority of studies to date have focused on metal contamination in freshwaters, the bioaccumulative capabilities of *A. aquaticus* and its effectiveness as a biomonitor. Most studies have focused on a subset of metals, including Cd, Cu, Zn and As, but Platinum-Group Elements (PGEs), such as Pt, Pd and Rh have also been examined due to prevalence in road-runoff, and there has been some work performed on studying the accumulation of Fe, Hg, Pb and Al in *A. aquaticus* (outlined in *Table 1*).

A previous review by (Goodyear and McNeill, 1999) summarised reports of the bioaccumulation of Zn, Cu, Pb and Cd in a number of aquatic macroinvertebrates, while a more recent review by (Ruchter et al., 2015) gave an overview of the effects of PGEs and their fate in the environment. This review aims to examine the current body of knowledge pertaining to the benthic isopod, *A. aquaticus*, with particular attention to the species' bioaccumulation of trace metals and possible explanations for its resilience to pollutants, as well as making an effort towards the assessment of its suitability as a biomonitor for environmental pollution.

Section II will introduce *Asellus aquaticus,* while drawing a contrast between the resilience of the species in the presence of pollutants to that of other aquatic macroinvertebrates. Section III will review the current understanding of potential and proposed mechanisms that allow for bioaccumulation of pollutants in *A. aquaticus*. Section IV will then discuss the possible use of *A. aquaticus* as a biomonitor, with particular attention to practical issues which may be encountered, before conclusions are given in Section V.

II. *Asellus aquaticus*

Asellus aquaticus is a freshwater isopod. Widely distributed throughout much of Europe and North America, commonly associated with a temperate climate, it has been recorded as far south as the Mediterranean, and as far north as Scandinavia (Maltby, 1991). The species is primarily associated with the β-mesosaprobic waters of the palearctic, which is an area defined by high concentrations of dissolved oxygen, low oxygen consumption and significant mineralisation of organic materials with end-products such as nitrates (de Nicola Giudici et al., 1988). *A. aquaticus* is known to feed primarily on decaying vegetation, microscopic algae and small invertebrates (Graça et al., 1993; Moore, 1975; Rossi and Fano, 1979). Several subspecies have been discovered, including *Asellus aquaticus infernus*, found in Romania, *A. a. cavernicolus*, *A. a. aquaticus* and *A. a. carniolicus*, all found in Slovenia (Sket, 1994; Sworobowicz et al., 2015; Turk et al., 1996; Turk-Prevorčnik and Blejec, 1998).

A. aquaticus is abundant across much of its range, and this distribution has rarely been influenced by anthropogenic impacts (Verovnik et al., 2005). It is tolerant to poor water quality and organic pollution (Maltby, 1995). It is, however, sensitive to sustained high temperatures, which has a negative effect on rates of growth, survival and reproduction (Di Lascio et al., 2011).

There has been some debate about the life cycle of the species, with a number of different breeding periods recorded, namely, winter breeding, summer breeding and year-long breeding. It is suggested that *A. aquaticu*s may completely replace two generations in one year, with the first overwintering generation breeding and dying in spring or early summer. The second generation grows, breeds and dies during the summer and is followed by the third generation, which survives the next winter, to again breed in the spring (Chambers, 1977). This would explain why both summer breeding and winter breeding have been observed. It has been suggested that the autumnal reproductive stasis may be explained by the lesser number of daylight hours during that period, and this leads to avoidance of breeding during less favourable, lower temperatures (Økland, 1978). One study noted that the overwintering spring individuals had a faster relative growth rate than the smaller individuals of the second generation (Adcock, 1979).

The size of an adult individual varies greatly over the species' range. Males of the species are generally found to be larger than females (Adams et al., 1985). There has been some suggestion that the size of *A. aquaticus* is related to the environment in which it lives, as individuals from clean water sites tend to be larger than those from polluted sites (Maltby, 1991; Tolba and Holdich, 1981), and, similarly, smaller individuals may be present in locations with higher temperature readings (Aston and Milner, 1980). A link may be drawn between tolerance of pollutants and the size of the individual (Kiffney and Clements, 1996). It is, therefore, important to take specimen size into account when gathering samples.

Benthic macroinvertebrates are particularly at risk to freshwater pollution, as they are in constant contact with both the sediment and water. Differing species show varying degrees of sensitivity and tolerance to various types of pollutants, with measurable responses ranging from growth inhibition to population die-off. It is by this end that a number of biotic indices have emerged, whereby the relative abundance of a species with a known tolerance to a pollutant can be used to extrapolate the severity of the pollution event (MacNeil et al., 2002). Biomonitoring studies including *A. aquaticus* as part of a wider cohort of biomonitor species have been carried out over a wide geographical and temporal range (Bascombe et al., 1990; Cao et al., 1996; Ketelaars and Frantzen, 1995).

One such example is the *Gammarus*:*Asellus* (G:A) ratio. The G:A ratio is based on the observation that *Gammarus pulex* and *Asellus aquaticus* frequently appear at the same location, but *G. pulex* is significantly less tolerant to organic pollution than *A. aquaticus*. If *A. aquaticus* is abundant at a site, but *G. pulex* not, the water at that site is thought to be polluted beyond the tolerance limit of *G. pulex* (Whitehurst, 1991). This index offers a simple, quick method of determining the severity of organic pollution in a freshwater system, however, the success of this method is very much dependent on the presence of both species in a river system (MacNeil et al., 2002). It should be noted that the difference in abundance between *G. pulex* and *A. aquaticus* may, alternatively, be due to competition between the two species, and that it may be as a result of this that they generally occur at different zones in a river system (Graça et al., 1994).

Similarly, *A. aquaticus* has been found to be more tolerant than other macroinvertebrate species to various types of pollutants. It was determined that the species is more tolerant than the amphipod, *Crangonyx pseudogracilis*, to inorganic pollution in all but the case of Al^{3+} and Mn^{2+} (Martin and Holdich, 1986). Another study compared *A. aquaticus* to *Ephoron virgo* and *Chironomus riparius*, and notes that of these three species, only *A. aquaticus* is limited to feeding from the sediment, while *E. virgo* and *C. riparius* can alter between sediment and water feeding (De Lange et al., 2005). This could greatly influence the types of pollutants *A. aquaticus* is exposed to.

III. Potential mechanisms

While many studies have investigated the effects of freshwater pollution on *A. aquaticus*, there remains a lack of firm understanding of the theoretical basis behind these results. Here, we attempt to provide an overview of the various proposed pathways of trace metal pollutants. We begin by studying the different forms in which pollutants can be present in the vicinity of the isopod, and the manner in which processes in the freshwater environment produces each of these forms. We then consider the proposed mechanisms by which *A. aquaticus* deals with the pollutants it encounters, and the subsequent fate of these pollutants. These mechanisms are initially studied under the single-element assumption, and we then consider the potential interactions between pollutants in the environment, and the manner in which this may impact upon the uptake and sequestering of these pollutants in *A. aquaticus*. Finally, we consider other stressors, and the indirect effects pollutants may have upon *A. aquaticus* via these stressors.

i. Pollutants in the environment

In order to quantify the pollutants that are bioavailable to benthic isopods, it is necessary to consider both the types of pollutants initially introduced to the environment, as well as the numerous processes which may transform the nature of the pollutants present in the environment.

Pollutants enter the freshwater environment via a number of pathways, which include, but are not limited to, domestic and industrial outflow, road run-off, agricultural run-off and natural leeching from mineral-rich soil. The nature of the pollutants entering the environment is not known *a priori*, but it must be assumed that the elements could be found in any state. Metal pollutants in freshwater systems may undergo precipitation or adsorption to suspended particles, which are then deposited as sediments (Atkinson et al., 2007). The metals can be present in very small concentrations, which makes quantification, or, in many cases, even detection, very difficult, but the effects are no less detrimental to the ecosystem, due to the potential for bioaccumulation and biomagnification.

However, environmental concentrations are not necessarily equivalent to concentrations bioaccumulated in the organism. Various processes can occur which may modify the state of pollutants once they have entered the environment. As outlined in *Fig. 2*, a number of mechanisms can take place, which may have an effect on the bioavailability of a metal species in the freshwater environment.

Metal speciation depends on a number of physicochemical factors such as pH, temperature, dissolved oxygen content and the availability of nitrogen- and sulphur-containing ligands with which the trace metals may form complexes which are biologically compatible. All of these factors, and more, determine the ultimate fate of the metal species in the aquatic environment, as they influence the bioavailability and toxicity of the metal (Kuppusamy and Giridhar, 2006; Pagnanelli et al., 2003).

There is also an indication that pH plays an important role in the uptake of metal species. While the common assumption is that a decrease in pH correlates to an increase in metal uptake, there have been a number of studies that contradict this, having seen an increased uptake under neutral conditions, compared to a lower pH (Gerhardt, 1993). The authors identify several possible explanations:

(a) At low pH, H^+ and the metal ion may compete for binding sites and/or the sensitivity of the carrier molecules for some metals may be lowered.

(b) At higher pH, metals may be adsorbed or co-precipitated and therefore less bioavailable.

(c) Surface adsorption onto the organism may account for increased metal levels at neutral pH.

(d) The organisms used for the experiment may have been from a site with pH neutral water, with the lowering pH resulting in stress and less ingestion by the organisms.

A number of other studies have given consideration to the correlation between pH and metal uptake (Campbel and Stokes, 1985; Courtney and Clements, 2000; Kar et al., 2008; Sako et al., 2009), but there remains much demand for further investigation.

pH dictates the cation and anion partitioning between the solid and liquid phases, and has an effect on the behaviour of the metal, meaning it may alter properties such as the bioavailability or toxicity of the metal (Warren and Haack, 2001).

A. aquaticus is unusual among benthic macroinvertebrates, as it has a significant salinity tolerance which may facilitate an extension of its range into brackish conditions (Lagerspetz and Mattila, 1961; Lockwood, 1959; Wolff, 1973). This raises the potential for its use as a biomonitor in both freshwater and estuarine systems. However, there is some debate about the mechanics of metal speciation in seawater, as opposed to freshwater. Although salinity is generally thought to influence the speciation of metals in water, one study found little difference for Cu, Pb or Zn between the freshwater and estuarine sections of a river, but there was significant reduction of Fe between the two sections (Hart and Davies, 1981). The oxidation characteristics of Fe have been found to differ in seawater and freshwater, and Fe has been seen to display a different pH dependency in the two water types (Hatje et al., 2003). This is attributed to rates of complexation and rates of oxygenation, rather than salinity. Care must, therefore, be taken not to freely extrapolate freshwater findings to the estuarine situation.

Other factors may also influence the speciation of metals, including weather, or even the presence of benthic dwellers. A significant relationship between metal speciation and heavy rainfall events has been observed, as the increased flow rate and load causes metal-containing sediments to become re-suspended (Meylan et al., 2003). Similarly, the disturbance of sediment by organisms is responsible, although at a small scale, for the re-suspension of metals (Atkinson et al., 2007).

Speciation distinguishes between free metal ions in solution, complexed metals in solution (which may be organic or inorganic in nature), adsorbed particles and colloidal particles. Furthermore, size fractionation differentiates between particles of various sizes.

Particles are measured on a micrometre (μm) scale, and larger sized particles may be removed from solute by way of filtration. However, particles smaller than the pores of the filter (typically 0.45μm) are referred to as dissolved fractions (Tuccillo, 2006). While it is not possible to remove these particles by simple filtration, a number of methods have been proposed for their removal, including sorption of the particles within a biofilm (Späth et al., 1998; Veglio and Beolchini, 1997). The size of a particle could be of great significance in the context of macroinvertebrates, although studies to test this have been inconclusive (Peeters et al., 2000), emphasising the need for subsequent trials.

Free metal ions are thought to be the most reactive species in freshwater, which implies greater toxicity, and, thus, a more urgent need for understanding (Allen et al., 1980). The free ion activity model (FIAM) has been put forward as a tool for assessing potential metal-organism interactions, and for determining the uptake and toxicity of all cationic trace metals. However, the FIAM makes the assumption that the activity of the free ion is directly related to the reactivity of the chemical species (Brown and Markich, 2000). The need for further studies based on field data has been identified, as the FIAM, while useful in a laboratory setting, is theoretical, and its applicability to real-world scenarios must be validated (Meylan et al., 2004). Despite efforts to examine the relationship between metal speciation and humic substances (Mantoura et al., 1978), there remains a need for practical studies to fully understand these interactions (Tipping et al., 2002).

A solid understanding of these various processes and mechanisms is vital to validating the implementation of *A. aquaticus* as an accumulative biomonitor. Ultimately, the issue of the interaction of organisms and pollutants in the environment cannot be answered solely through direct analysis of the environmental contamination. It would appear that two approaches are possible. Firstly, determination of the physicochemical and mechanical processes that the contaminants undergo, as well as the mechanics of bioavailability, would allow for the prediction of the potential threat to aquatic biota from pollution events. However, current understanding is insufficient to accurately predict the eventual speciation and distribution of pollutants in a waterway, partly due to the sheer complexity of the problem. The alternative approach would be to develop a means of assessing the bioavailable concentrations of pollutants. An accumulative biomonitor is ideal for this purpose, as it allows for the determination of pollutants at the point at which they enter the biosphere, in ignorance of the processes that the pollutants have undergone up to this point. In response to this, *A. aquaticus* has been the subject of various studies investigating the species' response to a variety of priority pollutants.

ii. Isolated pollutant uptake, sequestering & excretion in Asellus aquaticus

Presently, we must consider the proposed mechanisms of metal uptake in the context of isolated metal pollutants. Two potential pathways have been proposed for this sequestering of metals; ingestion (via the sediment), and surface adsorption (via both the water and sediment).

It is generally accepted that trace metals settle and accumulate in riverbed sediment, which leaves benthic organisms particularly exposed, and it is plausible that the uptake of metals by *A. aquaticus* could occur by way of an ingestion mechanism. Several authors suggest this pathway, based on measured high concentrations of PGE in the sediment, and trace concentrations in the water (Moldovan et al., 2001; Rauch and Morrison, 1999), and other studies also generally recognise ingestion as a likely route of metal uptake (Santoro et al., 2009).

However, one study found that there was a greater accumulation of Al by dead specimens than that of live specimens of *A. aquaticus*, which would seem to be in direct contradiction of the suggestion that the uptake of metals occurs by way of ingestion, and favours the theory of uptake by surface adsorption (Elangovan et al., 1999). Somewhat in keeping with this, water has been identified as the primary route for the uptake of ^{14}C-terbutryn and ^{14}C-benzo[*a*]pyrene (Richter and Nagel, 2007). Rather interestingly, another study found benzo[*a*]pyrene to have no observable effect on *A. aquaticus*, and also noted that varying the size of the organic particles made little difference to the uptake (Peeters et al., 2000).

It should be noted, then, that attempting to rule for one uptake mechanism over the other may be misguided, as it is very possible that both mechanisms occur to a significant level, under different conditions. The bioavailability of metals may vary greatly with complexation and concentration. With that in mind, the rate of uptake will likely differ greatly with each pathway and set of environmental conditions.

An interesting parallel may be drawn with findings that dietary uptake of hydrophobic pollutants increase with log K_{ow}, while aqueous uptake decreases with log K_{ow}, where K_{ow} is the octanol-water partition coefficient (Fisk et al., 1998; Qiao et al., 2000).

The mechanism by which bioaccumulation may occur has also been studied, but no solid conclusions have been drawn, due to the inherent difficulties involved in studying internal biological processes. However, two plausible theories emerge.

Metallothionein is linked to a metal capture-and-transport mechanism, and is associated with Zn regulation (Rauch and Morrison, 1999). The exact process by which this operates is unknown, but it is presumed that any binding to other metals involves Zn as an intermediate. Metallothionein has been studied in a range of species, not limited to crustaceans, and it should be noted that it has a high binding affinity for not just Zn (10^{-18}), but also Cd (10^{-22}). One theory suggests that the protein is present in invertebrates to regulate the uptake of Zn and Cu, and that it is by coincidence that it is also able to effectively regulate Cd (Talbot and Magee, 1978). There is still much debate about the exact mechanism by which metallothionein is able to protect the body against Cd toxicity, but there is consensus that it does (Klaassen et al., 1999).

In addition, it has been suggested that different metals would have different affinities to metallothionein, and would, therefore, differ in their ability to accumulate, or, indeed, the likelihood of accumulating (Berandah et al., 2010). To this end, greater accumulation of Pd and Pt compared to Rh, to which metallothioneins are known to have a lower affinity, has been observed (Moldovan et al., 2001), and an increased uptake rate of Pt (IV) over Pt (II) has been noted (Rauch and Morrison, 1999). Therefore, it would seem that *A. aquaticus*, for one reason or another, may be selectively accumulating metals.

On that note, it is worth specifying that the bioconcentration factor (BF) between *A. aquaticus* and the sediment has been seen to vary according to pollutant. For instance, one study found that the BF for Cd and Pb were 0.4 and 0.1, respectively, while that of Pt was found to be in the range of 4.8-28.6, which may suggest that *A. aquaticus* is better suited to the biomonitoring of some pollutants over others (Rauch and Morrison, 1999). Meanwhile, another study calculated the BF of Pd, Pt and Rh, as 150, 85, and 7, respectively (Moldovan et al., 2001). There is a clear discrepancy, in this case, between the two calculated BF values for Pt, for which the reason is unclear. There doesn't appear to have been much consideration for the differing contributions of water and sediment, both of which a benthic macroinvertebrate is in near-constant contact with. Since the sediment and water will likely have different concentrations and uptake mechanisms, this would justify the determination of differing BFs for each medium. Further investigation is needed in order to solidify our knowledge of the bioconcentration of various pollutants in *A. aquaticus*, and it is of great importance for the evaluation of a biomonitoring system that the various forms of a metal, as well as field conditions are taken into account.

Alternatively, it has been suggested that metals which have not been metabolized may accumulate in the digestive system (Köhler, 2002). In support of this theory, large, globular accumulations of Cu have been found in the hepatopancreas of *A. meridianus* (Brown, 1977), and such accumulation could very well occur in other species.

It would appear that the accumulation of metals often occurs in the exoskeleton of invertebrates, or in the soft tissue directly beneath the exoskeleton. The accumulation in *A. aquaticus* appears to consist of Type B granules in S-type cells (Hare, 1992), and 22x higher concentration of trace metals in a moulted carapace than in the rest of the body has been measured (Rauch and Morrison, 1999). As moulting the carapace would remove likely these harmful chemicals from the body, this could be an effective means of excreting harmful toxins, in which case, a study of the moulting habits and bioaccumulation sites in less-tolerant invertebrate species should show a differences.

In contrast, it has been suggested that *A. aquaticus* consumes its shed cuticle (Elangovan et al., 1999), which would result in re-accumulation of the metals, and a decrease in moulting frequency has been observed in the presence of Bisphenol A (BPA) (Plahuta et al., 2015), and again in the presence of wastewater samples (Plahuta et al., 2017), potentially due to endocrine disruption, which may also occur in heightened metal concentrations (Georgescu et al., 2011). Additionally, sewage effluent has been seen to inhibit the ventilation rate of *A. aquaticus*, which is an indication of reduced enzyme (cholinesterase and carboxylesterase) activity (O'Neill et al., 2004).

In one study, a number of static toxicity tests were carried out for the tolerance of *A. aquaticus* to various metal species, and it was found that the species was highly sensitive, although in varying degrees, to all metals tested (Migliore and de Nicola Giudici, 1990). Similarly, although not in keeping with the majority of the current body of literature on the subject, one study found *A. aquaticus* to be highly sensitive to Cu, and that the species is among the most sensitive macroinvertebrates to Cu to be studied as of yet (de Nicola Giudici et al., 1987). Further investigation is recommended.

Stable or radioisotope tracing could prove very useful in determining the ultimate fate of metals in the body of *A. aquaticus*. This would consist of labelling metals to render them detectable, and feeding them into the system via the suggested potential pathways. Making use of this method to determine the pathway through which uptake of Cd occurs in an isopod, one study found the Cd uptake to primarily occur through the water, and, therefore, suggested that increasing the metal partitioning to the sediment would result in a decreased bioaccumulation of Cd in the specimen (Eimers et al., 2002). Interestingly, it has been hypothesised that tolerance to Cd in *A. aquaticus* may occur if embryos are exposed to sub-lethal concentrations, prompting the tissues to develop necessary metal-binding proteins in order to overcome future Cd toxicity (Green et al., 1986).

iii. Synergistic & antagonistic effects

The majority of studies of trace metal pollutants have thus far focused on single metals and, as a result, have oversimplified potentially complex interactions between two or more metal species (Boyd, 2010). Consideration must be given to a scenario where a mixture of metals is present in the environment. There is much scope for further study in this area, as the examination of individual pollutants in isolation is not sufficient to offer insight into the mechanics of a real-world pollution event. One point that has been widely debated is the identification of whether a combination of metals has antagonistic or synergistic effects. This has previously been referred to as "additive", "more-than-additive" or "less-than-additive" (Pagenkopf, 1983), but for the purposes of this review, we have adhered to the more common terms of "antagonistic" and "synergistic" to describe to relationship between metals.

Antagonistic interactions may occur when two or more metals have similar properties, and compete for priority binding. This most readily occurs in the case of protein binding sites. While the sites selectively favour an element over others, it can occur that more than one element will fit the criteria for binding to a specific site. For example, one study predicted that Cd and Pb both compete for Ca^{2+}

binding sites, so one would expect to see that those species will have antagonistic interactions (Van Ginneken et al., 2015).

With that in mind, there are a number of properties that may give an indication of the likelihood of correctly predicting which metal in a mixture is most suited to binding, such as the co-ordination number (CN) of the metal, or the co-ordination geometry. Other factors include the valence state and ionic radius of the metal, as well as the charge and polarizability of the ligand. All of these factors influence the potential mechanism, and a thorough examination of the nature of the metal species may help to determine the effects of each species in a mixture. However, as of yet, we do not have a complete understanding of the process by which metallothionein selects the required metal.

That said, some exploratory work has been carried out into the accumulation of metal mixtures by *A. aquaticus*. It is apparent that Zn^{2+} is predisposed to competition from Cu^{2+}. Also noteworthy from one paper is the authors' statement that a comprehensive understanding of the competition between biologically-necessary metals and toxic metal species is lacking (Dudev and Lim, 2013). The authors suggest that further investigation is needed in the following areas:

(a) factors controlling the kinetics of the metal exchange in protein binding sites;
(b) the effects of ionic strength on the process and selectivity;
(c) the biological role of various metal ions;
(d) thorough studies of the factors influencing anion selectivity.

Other factors may influence the interactions between metals. It has been tentatively established that there is a strong relationship between pH and metal contaminant uptake in sediment (Sako et al., 2009), and it has also been suggested that selective binding is potentially linked to increased body water content (Hargeby, 1990), while others argue that it may be due to metabolic potential (Simčič and Brancelj, 2006). Additionally, the intrinsic properties of each of the elements will factor in determining which mechanism is more plausible. While the metal ion will compete with other metallic cations in the environment, certain evolutions must have taken place to ensure that a specific metal will bind to the protein, as may be required by the creature's body (Dudev and Lim, 2013). This refers to the evolution of the cell, drawing the conclusion that metalloproteins developed selectivity for certain metals due to an abundance of those species in the primordial ocean from which the cell evolved.

One study determined that a significant percentage of interactions between Cd, Cu and Zn, in the freshwater environment, are antagonistic in type. Additionally, the point was emphasised, that where the specimen is exposed to a mixture of contaminants, the outcome is difficult to predict, and models thus far have been somewhat imprecise. The authors of that paper go one step further, and state that current scientific understanding does not yet allow for the possibility of predicting the outcome when multiple contaminants are present, and the best we can hope to achieve is to identify a probable outcome for certain types of cases (Vijver et al., 2011).

However, elucidation of the differences between isolated exposures and complex mixtures has only just begun, and a more complete understanding would be required before the biomonitor is considered suitable for use in a practical setting

iv. Other stressors in the environment

There are many factors, other than the presence of metal pollutants, which could influence the survival and health of *Asellus aquaticus*, many of which have been investigated in-depth. Most of these parameters are physicochemical in nature, and while it is difficult to consider every possible scenario, it is of the utmost importance that at least some thought is given to natural conditions.

Temperature is one such parameter that has been studied in the context of *A. aquaticus*, and it has been determined that the survival rate of the species is reduced at high temperatures (Ieromina et al., 2014). It has also been found that the survival rate of eggs and embryos is highest at lower temperatures (Holdich and Tolba, 1981). It has been found that the optimal temperature for the embryo development of *A. aquaticus* is in the range of 14.5 – 18.8ºC (Roshchin and Mazelev, 1979). *A. aquaticus* has been observed using defence mechanisms against high temperatures, namely behavioural avoidance reactions by way of increased turnings, or "klinokinesis", induction or increase in production of a heat shock protein associated with an increase in thermal tolerance, and the slower but longer-lasting mechanism of thermal acclimation, associated with CNS and/or endocrine effects (Lagerspetz, 2003).

It has been proposed that the Fluctuating-Asymmetry (FA) observed when *A. aquaticus* is subject to temperature-related stress could be employed as a measure of overall environmental stress (Savage and Hogarth, 1999). Although this method would be low-cost, quick and require minimal formal training on the part of the observer, it is not clear if the sample size used in the study was sufficient to definitively draw a parallel between a temperature stressor and FA observed. As it is possible that other stressors may not have been fully accounted for, further investigation would need to be carried out before this method could be accepted as an indicator for general environmental health.

Dissolved oxygen levels are also known to have an influence on the growth of *A. aquaticus* (Maltby, 1995). As one would expect, this has been confirmed by a number of studies, with the observation of impaired growth of the organisms in a low oxygen environment. Growth inhibition is associated with a higher energy requirement for respiration (Ieromina et al., 2014).

On this point, it is necessary to note that other authors have held the assumption that *A. aquaticus* is tolerant of low oxygen conditions (De Smet and Das, 1981), so further investigation is recommended in order to determine the species' limits of tolerance to this parameter.

A change in the pH of a stream can have a detrimental effect on a large number of species. Acidification may also play a part in the health of *A. aquaticus*, as has been outlined in a number of studies. A reduction in the ingestion rate of *A. aquaticus* was seen as acidification increased (Costantini et al., 2005). While it has been found that *A. aquaticus* is affected by acidification, some authors note that the species is more tolerant than other crustacean species, namely *Gammarus pulex*, as well as noting that specimens from polluted water sites may be more tolerant to acidification than specimens from clean water sites (Naylor et al., 1990).

Despite the general concern about pesticides entering the hydrosphere (Rasmussen et al., 2015), they do not seem to pose a direct threat to *A. aquaticus*. It is unclear by what mechanism the species has developed a tolerance to pesticide pollution, but a number of studies have verified this claim (Bundschuh et al., 2012; Ieromina et al., 2014; Lukančič et al., 2010). Once again, however, the literature is contradictory, as, in a study of various species' sensitivity to an insecticide, it was found that *A. aquaticus* had a tolerance almost as poor as the tested insect species, and was found to be the most sensitive of any crustacean species tested (Finnegan et al., 2018). This again emphasises the need for methodical testing of the species to a range of pollutants, under various conditions, to determine the tolerance.

While most authors believe that pesticides do not appear to harm *A. aquaticus*, the use of fungicides may limit the amount of fungal growth on leaves, thus reducing the availability of food for *A. aquaticus*, and, in turn, having an effect on the growth and reproduction of the species (Feckler et al., 2016; Gardeström et al., 2016).

Another factor to consider is the presence of other organisms in the vicinity of *A.* aquaticus, as interactions with other species may be antagonistic. The species is commonly predated upon by a range of visual predators, which, it has been suggested, is the driving force behind the evolution of a transparent, or cryptically pigmented, carapace (Hargeby et al., 2005). It is known to be predated on by a number of species, including fish (Dallinger and Kautzky, 1985; Rask and Hiisivuori, 1985), flatworms (Bundschuh et al., 2012; Ham et al., 1995), amphibians (Verrell, 1985), and other crustacean species (Dick et al., 2002). Interestingly, it has been found that *A. aquaticus* is most frequently predated upon by perch at dawn and dusk (Persson, 1983).

There has been some suggestion that the precopulatory mate guarding mechanism of *A. aquaticus* has evolved in response to predation, in order to maximise survival and mating success (Benesh et al., 2007; Verrell, 1985). Furthermore, the fitness and survival of *A. aquaticus* is said to decrease in the presence of *Gammarus pulex*, potentially attributable to the prolonged excretion of amino acids as a chemical avoidance strategy used by *A. aquaticus* following visual or chemical contact with *G. pulex* (Blockwell et al., 1998).

A. aquaticus may also be subject to parasitism, with finding that suggest that juveniles and maturing adults exposed to parasites may have a greater survival rate than adults exposed (Hasu et al., 2006). At least one study has found that the parasite, in this case *Acanthocephalus lucii*, also contained Cd and Pb concentrations, although significantly lower than that found in the host (Sures and Taraschewski, 1995).

There are numerous other factors effecting the growth, health and survival rate of *A. aquaticus*, and although it would be difficult to account for all possible stressors, it is important that a potential biomonitoring scheme should try to account for as many factors as possible, and it does not seem outrageous that said programme should at least, in order to be thorough in its investigation, include routine measurements for parameters such as temperature and pH.

IV. Use of *Asellus aquaticus* as a biomonitor

A biomonitor is a species which exhibits a quantitative response to the presence of a pollutant in the environment. An effective biomonitor has the following qualities (Johnson and Wiederholm, 1993; Usseglio-Polatera et al., 2000):

(a) a measurable reaction occurs in the presence of the pollutant of interest;
(b) the reaction is relatively easy to measure or observe;
(c) it is abundant and widespread;
(d) it can tolerate the conditions in which the measurement is to take place;
(e) a change occurs in the reaction metric over a wide range of pollutant concentrations;
(f) the reaction is insensitive to other factors, or can be calibrated against other factors.

A biomarker is a specific type of qualitative biomonitor which exhibits a binary response, and is, therefore, only useful in determining the presence, but not the concentration, of a pollutant (Ansari et al., 2008; de la Torre et al., 2001).

One could potentially suggest many macroinvertebrates which may serve as effective biomonitors of freshwater pollution, however, there are several characteristics specific to *A. aquaticus*, including some aspects of the life history of the species, which give the isopod a number of advantages over other macroinvertebrates, identifying it as a particularly suitable candidate.

With its wide geographical range and abundance, *A. aquaticus* has long been of interest to freshwater ecologists, and has frequently been utilised, alongside other macroinvertebrates, as a bioindicator of water quality, as well as in the determination of the way in which a given benthic community may respond to changes in the ecosystem over time, due to pollution (Bascombe et al., 1988; Bergfur et al., 2007; Hunting et al., 2013; Montañés et al., 1995). *A. aquaticus*, as a relatively common macroinvertebrate which can be found in most temperate freshwater systems, has been the subject of a variety of studies of metal toxicity and accumulation for several decades (Canivet et al., 2001; Green et al., 1986; Van Ginneken et al., 2015). Significantly, the organism cannot swim, and often lives quite a localised existence within the sediment of water bodies, which, in that respect, makes it an ideal candidate for spatial biomonitoring (Resh, 2008).

Although *Asellus aquaticus* has been proposed as a biomonitor in a number of studies, it is worth pointing out that there have been both successful and unsuccessful applications of this theory.

Previous studies have examined the use of *A. aquaticus* as a biomonitor for a number of common pollutants. The effect of Cu and As on other reactions (Na^+/K^+-ATPase, Metallothionein and TBARS) has been studied, and an increase in reactivity with regard to the latter two mechanisms when these metals are in the environment was observed, indicating that *A. aquaticus* can be used as a biomarker by examining the effects of these reactions, and therefore the reactivity (Bouskill et al., 2006).

One study, however, should serve as a warning of the potential for complications in applying *A. aquaticus* as a biomonitor (Kaya et al., 2014). No significant differences in Na^+/K^+-ATPase or TBARS were noted in the presence of high levels of trace metals; however, the authors noted that there was an increase in total glutathione levels, which is indicative of reactive oxygen species production. They suggest that the organisms have adapted to the polluted environment by producing antioxidants to combat the increased oxidative stress. Further studies should be carried out to determine if this could lead to a reduction in bioassay effectiveness over time.

An example of a full-scale biomonitoring project with mixed results has been described, wherein a long-term study with temporal and spatial dimensions involving a number of biomonitor species, including *A. aquaticus*, failed to show statistical trends for any of the species studied. It was suggested that reduced bioavailability of the metals could be at fault (Kolaříková et al., 2012).

One point that is worth emphasizing, and could prove crucial in the implementation of any subsequent biomonitoring programme, is the assumption made in many of these studies that superposition holds, that by studying the effects of each pollutant in isolation, we can predict how a real-world situation, with many different pollutants, will affect the animal.

Firstly, as shown by at least two studies, the biomonitor responds differently when more than one pollutant is present, potentially due to the various pollutants competing for the same binding site (Charles et al., 2014; Van Ginneken et al., 2015). Secondly, complexation may play a role, as there is no reason that a mechanism which would work for a pollutant in isolation should work similarly for grouped elements. This is one of the key points of this paper – the current way of approaching the problem is too simplistic.

There is a complex relationship between predator stress and stress due to the presence of metal pollutants, in isolation or in a mixture (Van Ginneken et al., 2018). It would appear, then, that a successful biomonitoring programme would need to take into account the sensitivity of the biomonitor mechanism to various stressors, other than those being measured.

During the design process of a typical biomonitoring programme, the response of the organism, in terms of a measurable quantity, N, is evaluated at multiple concentrations of the pollutant, C_x. These parameters are then used to estimate the conversion function of the model:

$$N=f'(C_x)$$

However, if other pollutants ($C_1, C_2, ...$) and/or other stressors ($S_1, S_2, ...$) are present, and have an effect on the response of the biomonitor, the resulting real-world conversion function is:

$$N=f(C_x, C_1, C_2, ... S_1, S_2, ...)$$

The estimated conversion function is, therefore, only valid under the stressor conditions under which it is evaluated. For this reason, *ex-situ* tests can only ever be considered as a crude approximation of *in-situ* conditions, unless a comprehensive range of factors, other than those of interest, have also been studied (Bloor and Banks, 2006).

For a more robust biomonitor, all factors which could significantly influence the conversion function must be identified and accounted for.

As has been explored in Section V, the mechanism of pollutant uptake by *A. aquaticus* is likely to be complicated, suggesting that there is a plethora of potential synergistic and/or antagonistic interactions between stressors. Some of these interactions may be unexpected, as is seen in one case where effects on an algal community were attributed to the response of the grazer community to the combination of elevated nutrient levels and a cocktail of metal contaminants (Breitburg et al., 1999).

It is likely that any specimens used for a biomonitoring programme would be more suitable if they were bred for the purpose, and placed *in-situ*, as this would ensure all specimens are of a similar age, and would help to avoid the inclusion of individuals which may have been exposed for a longer period, thus skewing the results. A study that examined the use of juvenile brown trout as potential biomonitors of metal pollution in freshwater backs up this argument, stating that it is useful to standardise the breeding of the organisms to a certain timeframe, as the accumulated pollutants would then clearly correspond to the contamination within that time period (Lamas et al., 2007).

When considering the use of *A. aquaticus* as a biomonitor, it should be noted that juvenile individuals bred from specimens which were subject to sub-lethal doses of pollutants have been found to be less sensitive than the parent generation (de Nicola Giudici et al., 1986). There is some suggestion that this adaption is genetically based, but this point has been challenged by one study, which determined that long-term isolation of a population had little effect, when compared to other populations, on the tolerance level that *A. aquaticus* had to Cd (Pascoe and Carroll, 2004).

Further incentive for the use of macroinvertebrates as part of a biomonitoring programme is that they are predated upon, and by way of this, the pollutants that have been bioaccumulated may enter into the food chain. It has been found that fish which feed on macroinvertebrates have a higher concentration of Cd and Zn, than piscovorous species (Amundsen et al., 1997).

The use of a species as a biomonitor is never clear-cut, as there is always debate about various properties and characteristics. For example, one paper makes the point that the most persistent compounds will be most abundant in the tissues of older organisms (Lamas et al., 2007). While this certainly makes sense in terms of bioaccumulation over a lifetime, another paper directly contradicts this statement by suggesting metal concentrations are lower in older specimens, as they may excrete the pollutant more efficiently than the juvenile specimens (Amundsen et al., 1997). Both parties make a compelling argument, and perhaps this varies with chosen species, but the point remains that there is never a single “turn-key” biomonitor species that can be used in any given scenario, and a great amount of investigation and understanding must take place before a programme can be successfully implemented.

V. Conclusions

This review, while by no means exhaustive, has outlined much of the general understanding, thus far, regarding *Asellus aquaticus*, and, more specifically, its use as a biomonitor for trace metal pollutants in freshwater systems. By way of reviewing the current and past literature pertaining to the subject, a number of points have come to light, and it follows that several areas or topics have much scope for expansion.

Although trace metals alone pose a threat to the environment, further thought must be given to how these pollutants may be altered or accelerated as a result of other incidental factors in a real-world setting. Further studies on the interactions between metals, and the consequences of such for benthic macroinvertebrates, are greatly needed in order to expand our understanding of the ways by which a mixture of metals may impact upon biota.

Our understanding of uptake, sequestering and excretion of metals by *A. aquaticus* is minimal, and any amount of further investigation in this area would greatly improve the current state of knowledge. Even better, would be expansion of the body of data pertaining to field samples, and the uptake of metals outside of a laboratory setting.

The *Gammarus:Asellus* ratio and the observation of Fluctuating-Asymmetry both hold great potential as techniques that could be implemented, perhaps even with a citizen science application, but are sorely in need of further validation in order to cement the theory, as there is significant argument which negates the assumptions put forward by the methods.

It would appear from the presented literature, that toxicity tests are a good format for the validation of the effects various pollutants have on *A. aquaticus*. However, a great number of common and exotic pollutants have yet to be tested, and there have not been enough replicates of those few that have been carried out. Too much emphasis has been put on a study being the first of its kind, and not nearly enough on replicating and validating existing results. It is only by doing this that a method may actually be rolled out for practical use.

In the case of *Asellus aquaticus,* one could make the argument that while there remains much to be understood about the species, and, in particular, the mechanism by which it sequesters metal pollutants, it has already been determined that the species is remarkably tolerant to poor water conditions and it has been ascertained that it does bioaccumulate metals. These points, as well as the wide geographical range, abundance and fast reproduction rate of the species make it, once some further validation has been successfully carried out, an ideal candidate for use in a biomonitoring programme for the detection and quantification of metal pollution.

Acknowledgements – The authors wish to acknowledge the support of the Irish Research Council and the Environmental Protection Agency (Ireland) under Grant No. GOIPG/2018/3.

Tables and Figures

Table 1: A selection of studies investigating the interaction between various metal species and Asellus aquaticus.

REFERENCE	ELEMENT OF INTEREST																								
	Al	K	Ca	Cr	Mn	Fe	Ni	Cu	Zn	As	Br	Rb	Sr	Y	Zr	Rh	Pd	Cd	Sb	Cs	Ba	W	Pt	Hg	Pb
Ekvall (2018)																						✔			
Van Ginneken et al., 2018								✔										✔							✔
Joachim et al., 2017								✔																	
Van Ginneken et al., 2017								✔										✔							✔
Palma et al., 2015				✔				✔	✔	✔								✔							✔
Van Ginneken et al., 2015								✔										✔							✔
Kaya et al., 2014						✔		✔	✔									✔							✔
Hunting et al., 2013								✔																	
Haus et al., 2007				✔				✔	✔									✔	✔				✔		✔
Bouskill et al., 2006								✔		✔															
Tulonen et al., 2006				✔														✔						✔	✔
Akyüz et al., 2001		✔	✔		✔	✔	✔	✔	✔		✔	✔	✔	✔	✔			✔		✔	✔			✔	✔
Canivet et al., 2001				✔						✔															
Moldovan et al., 2001																✔	✔						✔		
Elangovan et al., 1999	✔																								
Rauch and Morrison, 1999																							✔		
Van Hattum et al., 1996								✔	✔									✔							✔
Ham et al., 1995																		✔							
Sures and Taraschewski, 1995																		✔							✔
Migliore and de Nicola Giudici, 1990				✔		✔	✔	✔	✔									✔						✔	✔
de Nicola Giudici et al., 1988								✔										✔							
de Nicola Giudici et al., 1987								✔																	
Green et al., 1986																		✔							
Fraser, 1980																									✔
Fraser et al., 1978																									✔

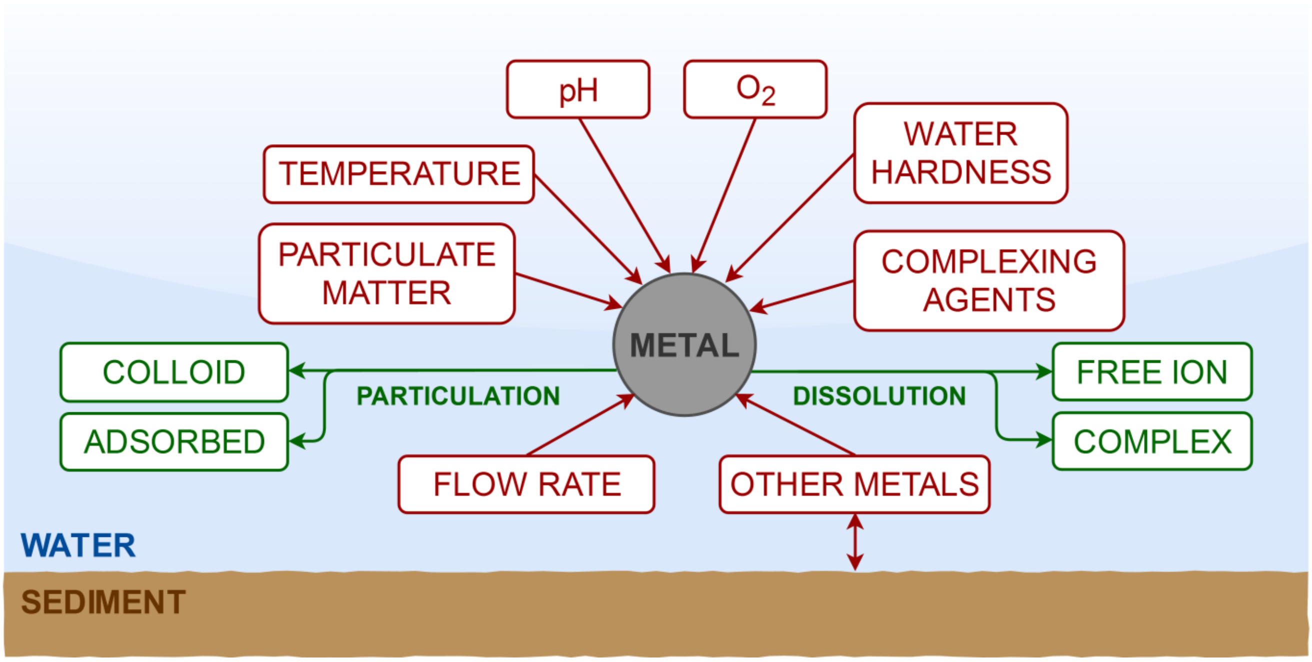

Fig. 1: Factors which influence metal speciation - from Gerhardt, 1993, modified.

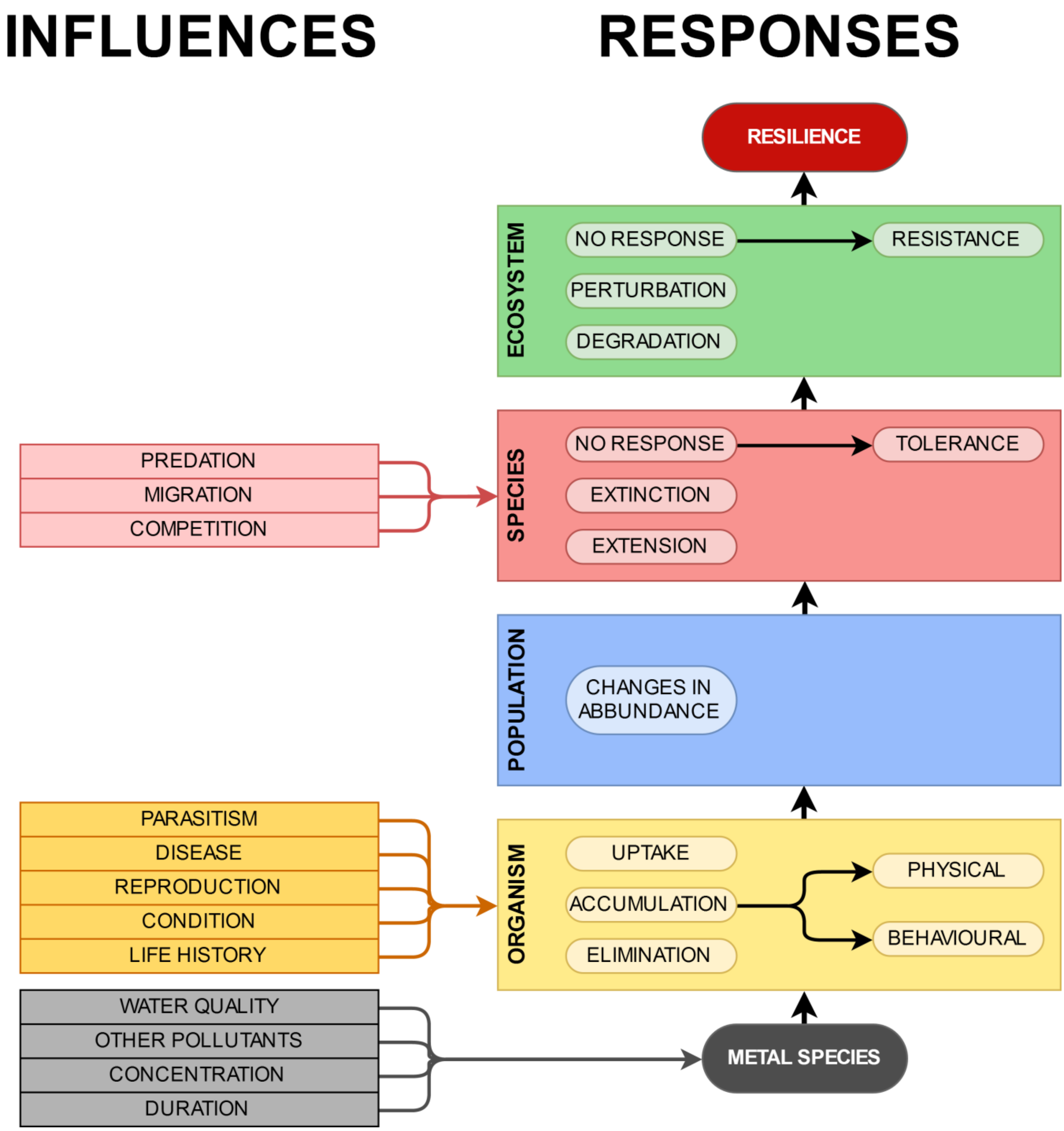

Fig. 2: An overview of responses of organisms, species and ecosystems to factors influencing them - from Gerhardt, 1993, modified.

References

Adams, J., Greenwood, P., Pollitt, R., Yonow, T., 1985. Loading constraints and sexual size dimorphism in Asellus aquaticus. Behaviour 277–287.

Adcock, J.A., 1979. Energetics of a population of the isopod Asellus aquaticus: life history and production. Freshwater Biology 9, 343–355.

Adriaenssens, V., Goethals, P.L., De Pauw, N., 2006. Fuzzy knowledge-based models for prediction of Asellus and Gammarus in watercourses in Flanders (Belgium). Ecological Modelling 195, 3–10.

Akyüz, T., Erkan, B.M., Akyüz, S., Bassari, A., 2001. Radioisotope excited X-ray fluorescence analysis of Asellus Aquaticus (Crustacea: Isopoda) from Istanbul as an indicator of environmental metal pollution. Journal of Radioanalytical & Nuclear Chemistry 249, 649–651.

Allen, H.E., Hall, R.H., Brisbin, T.D., 1980. Metal speciation. Effects on aquatic toxicity. Environmental science & technology 14, 441–443.

Amundsen, P.-A., Staldvik, F.J., Lukin, A.A., Kashulin, N.A., Popova, O.A., Reshetnikov, Y.S., 1997. Heavy metal contamination in freshwater fish from the border region between Norway and Russia. Science of the Total Environment 201, 211–224.

Ansari, R. A., Kaur, M., Ahmad, F., Rahman, S., Rashid, H., Islam, F., Raisuddin, S., 2009. Genotoxic and oxidative stress-inducing effects of deltamethrin in the erythrocytes of a freshwater biomarker fish species, Channa punctata Bloch. Environmental Toxicology: An International Journal 24.5, 429-436.

Aston, R.J., Milner, A.G.P., 1980. A comparison of populations of the isopod Asellus aquaticus above and below power stations in organically polluted reaches of the River Trent. Freshwater biology 10, 1–14.

Atkinson, C.A., Jolley, D.F., Simpson, S.L., 2007. Effect of overlying water pH, dissolved oxygen, salinity and sediment disturbances on metal release and sequestration from metal contaminated marine sediments. Chemosphere 69, 1428–1437.

Bascombe, A.D., Ellis, J.B., Revitt, D.M., Shutes, R.B.E., 1988. Macroinvertebrate biomonitoring and water quality management within urban catchments, in: Hydrological Processes and Water Management in Urban Areas (Proceedings of the Duisberg Symposium, April 1998).

Benesh, D.P., Valtonen, E.T., Jormalainen, V., 2007. Reduced survival associated with precopulatory mate guarding in male Asellus aquaticus (Isopoda), in: Annales Zoologici Fennici. JSTOR, pp. 425–434.

Berandah, F.E., Kong, Y.C., Ismail, A., 2010. Bioaccumulation and distribution of heavy metals (Cd, Cu, Fe, Ni, Pb and Zn) in the different tissues of Chicoreus capucinus lamarck (Mollusca: Muricidae) collected from Sungai Janggut, Kuala Langat, Malaysia. Environ Asia 3, 65–71.

Bergfur, J., Johnson, R.K., Sandin, L., Goedkoop, W., Nygren, K., 2007. Effects of nutrient enrichment on boreal streams: invertebrates, fungi and leaf-litter breakdown. Freshwater Biology 52, 1618–1633.

Blockwell, S.J., Taylor, E.J., Jones, I., Pascoe, D., 1998. The influence of fresh water pollutants and interaction with Asellus aquaticus (L.) on the feeding activity of Gammarus pulex (L.). Archives of Environmental Contamination and Toxicology 34, 41–47.

Bloor, M.C., Banks, C.J., 2006. An evaluation of mixed species in-situ and ex-situ feeding assays: The altered response of Asellus aquaticus and Gammarus pulex. Environment International 32, 22–27.

Bouskill, N.J., Handy, R.D., Ford, T.E., Galloway, T.S., 2006. Differentiating copper and arsenic toxicity using biochemical biomarkers in Asellus aquaticus and Dreissena polymorpha. Ecotoxicology and Environmental Safety 65, 342–349.

Boyd, R.S., 2010. Heavy metal pollutants and chemical ecology: exploring new frontiers. Journal of chemical ecology 36, 46–58.

Breitburg, D.L., Sanders, J.G., Gilmour, C.C., Hatfield, C.A., Osman, R.W., Riedel, G.F., Seitzinger, S.P., Seitzinger, S.P., 1999. Variability in responses to nutrients and trace elements, and transmission of stressor effects through an estuarine food web. Limnology and Oceanography 44, 837–863.

Brown, B.E., 1977. Uptake of copper and lead by a metaltolerant isopod Asellus meridianus Rac. Freshwater Biology 7, 235–244.

Brown, P.L., Markich, S.J., 2000. Evaluation of the free ion activity model of metal-organism interaction: extension of the conceptual model. Aquatic Toxicology 51, 177–194.

Bundschuh, M., Appeltauer, A., Dabrunz, A., Schulz, R., 2012. Combined effect of invertebrate predation and sublethal pesticide exposure on the behavior and survival of Asellus aquaticus (Crustacea; Isopoda). Archives of environmental contamination and toxicology 63, 77–85.

Campbel, P.G.C., Stokes, P.M., 1985. Acidification and toxicity of metals to aquatic biota. Canadian Journal of Fisheries and Aquatic Sciences 42, 2034–2049.

Canivet, V., Chambon, P., Gibert, J., 2001. Toxicity and bioaccumulation of arsenic and chromium in epigean and hypogean freshwater macroinvertebrates. Archives of Environmental Contamination and Toxicology 40, 345–354.

Cao, Y., Bark, A.W., Williams, W.P., 1996. Measuring the responses of macroinvertebrate communities to water pollution: a comparison of multivariate approaches, biotic and diversity indices. Hydrobiologia, 341, 1-19.

Chambers, M.R., 1977. A comparison of the population ecology of Asellus aquaticus (L.) and Asellus meridianus Rac. in the reed beds of the Tjeukemeer. Hydrobiologia 53, 147–154.

Charles, J., Crini, G., Degiorgi, F., Sancey, B., Morin-Crini, N., Badot, P.-M., 2014. Unexpected toxic interactions in the freshwater amphipod Gammarus pulex (L.) exposed to binary copper and nickel mixtures. Environmental Science and Pollution Research 21, 1099–1111.

Costantini, M.L., Mancinelli, G., Mandrone, S., Rossi, L., 2005. Combined effects of acidification and competition on the feeding preference of a freshwater macroinvertebrate, Asellus aquaticus (Crustacea: Isopoda): a laboratory experiment. Marine and Freshwater Research 56, 997–1004.

Courtney, L.A., Clements, W.H., 2000. Sensitivity to acidic pH in benthic invertebrate assemblages with different histories of exposure to metals. Journal of the North American Benthological Society 19, 112–127.

Dallinger, R., Kautzky, H., 1985. The importance of contaminated food for the uptake of heavy metals by rainbow trout (Salmo gairdneri): a field study. Oecologia 67, 82–89.

De Jonge, M., Van de Vijver, B., Blust, R., Bervoets, L., 2008. Responses of aquatic organisms to metal pollution in a lowland river in Flanders: a comparison of diatoms and macroinvertebrates. Science of the Total Environment 407, 615–629.

De Lange, H.J., De Haas, E.M., Maas, H., Peeters, E.T.H.M., 2005. Contaminated sediments and bioassay responses of three macroinvertebrates, the midge larva Chironomus riparius, the water louse Asellus aquaticus and the mayfly nymph Ephoron virgo. Chemosphere 61, 1700–1709.

De Lange, H.J., Sperber, V., Peeters, E.T.H.M., 2006. Avoidance of Polycyclic Aromatic Hydrocarbon-Contaminated Sediments by the Freshwater Invertebrates Gammarus pulex and Asellus aquaticus. Environmental Toxicology and Chemistry 25, 452.

de la Torre, F.R., Ferrari, L., Salibian, A., 2002. Freshwater pollution biomarker: response of brain acetylcholinesterase activity in two fish species. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology, 131, 271-280.

de Nicola Giudici, M., Migliore, L., Gambardella, C., Marotta, A., 1988. Effect of chronic exposure to cadmium and copper on Asellus aquaticus (L.)(Crustacea, Isopoda). Hydrobiologia 157, 265–269.
de Nicola Giudici, M., Migliore, L., Guarino, S.M., 1987. Sensitivity of Asellus aquaticus (L.) and Proasellus coxalis Dollf.(Crustacea, Isopoda) to copper. Hydrobiologia 146, 63–69.
de Nicola Giudici, M., Migliore, L., Guarino, S.M., 1986. Effects of cadmium on the life cycle of Asellus aquaticus (L.) and Proasellus coxalis Dollf.(Crustacea, Isopoda). Environmental Technology 7, 45–54.
De Smet, W.H.O., Das, A.M.L., 1981. Of A sellus aquaticus (L.) (Crustacea, Isopoda) in an Aerated Brook.
Di Lascio, A., Rossi, L., Letizia Costantini, M., 2011. Different temperature tolerance of northern and southern European populations of a freshwater Isopod Crustacean species (Asellus aquaticus L.). Fundamental and Applied Limnology/Archiv für Hydrobiologie 179, 193–201.
Dick, J.T., Platvoet, D., Kelly, D.W., 2002. Predatory impact of the freshwater invader Dikerogammarus villosus (Crustacea: Amphipoda). Canadian Journal of Fisheries and Aquatic Sciences 59, 1078–1084.
Dudev, T., Lim, C., 2013. Competition among metal ions for protein binding sites: determinants of metal ion selectivity in proteins. Chemical reviews 114, 538–556.
Eimers, M.C., Evans, R.D., Welbourn, P.M., 2002. Partitioning and bioaccumulation of cadmium in artificial sediment systems: application of a stable isotope tracer technique. Chemosphere 46, 543–551.
Ekvall, M.T., Hedberg, J., Odnevall Wallinder, I., Hansson, L.-A., Cedervall, T., 2018. Long-term effects of tungsten carbide (WC) nanoparticles in pelagic and benthic aquatic ecosystems. Nanotoxicology 12, 79–89.
El Gawad, S.S.I.A., 2009. The Mollusk Gastropod Lanistes carinatus (Olivier, 1804) as Abiomonitor for Some Trace Metals in the Nile River. International Journal of Zoological Research 5, 115–125.
Elangovan, R., Ballance, S., White, K.., McCrohan, C.., Powell, J.., 1999. Accumulation of aluminium by the freshwater crustacean Asellus aquaticus in neutral water. Environmental Pollution 106, 257–263.
Feckler, A., Goedkoop, W., Zubrod, J.P., Schulz, R., Bundschuh, M., 2016. Exposure pathway-dependent effects of the fungicide epoxiconazole on a decomposer-detritivore system. Science of the Total Environment 571, 992–1000.
Finnegan, M.C., Emburey, S., Hommen, U., Baxter, L.R., Hoekstra, P.F., Hanson, M.L., Thompson, H., Hamer, M., 2018. A freshwater mesocosm study into the effects of the neonicotinoid insecticide thiamethoxam at multiple trophic levels. Environmental Pollution 242, 1444–1457.
Fisk, A.T., Norstrom, R.J., Cymbalisty, C.D., Muir, D.C., 1998. Dietary accumulation and depuration of hydrophobic organochlorines: bioaccumulation parameters and their relationship with the octanol/water partition coefficient. Environmental Toxicology and Chemistry 17, 951–961.
Fleeger, J.W., Carman, K.R., Nisbet, R.M., 2003. Indirect effects of contaminants in aquatic ecosystems. Science of the Total Environment 317, 207–233.
Fraser, J., 1980. Acclimation to lead in the freshwater isopod Asellus aquaticus. Oecologia 45, 419–420.
Fraser, J., Parkin, D.T., Verspoor, E., 1978. Tolerance to lead in the freshwater isopod asellus aquaticus. Water Research 12, 637–641.
Fuller, N., Ford, A.T., Nagorskaya, L.L., Gudkov, D.I., Smith, J.T., 2018. Reproduction in the freshwater crustacean Asellus aquaticus along a gradient of radionuclide contamination at Chernobyl. Science of the Total Environment 628, 11–17.

Gardeström, J., Ermold, M., Goedkoop, W., McKie, B.G., 2016. Disturbance history influences stressor impacts: effects of a fungicide and nutrients on microbial diversity and litter decomposition. Freshwater biology 61, 2171–2184.
Georgescu, B., Georgescu, C., Dărăban, S., Bouaru, A., Paşcalău, S., 2011. Heavy metals acting as endocrine disrupters. Scientific Papers Animal Science and Biotechnologies 44, 89–93.
Gerhardt, A., 1993. Review of impact of heavy metals on stream invertebrates with special emphasis on acid conditions. Water, Air, and Soil Pollution 66, 289–314.
Goodyear, K.., McNeill, S., 1999. Bioaccumulation of heavy metals by aquatic macro-invertebrates of different feeding guilds: a review. Science of The Total Environment 229, 1–19.
Graça, M.A.S., Maltby, L., Calow, P., 1994. Comparative ecology of Gammarus pulex (L.) and Asellus aquaticus (L.) I: population dynamics and microdistribution. Hydrobiologia 281, 155–162.
Graça, M.A.S., Maltby, L., Calow, P., 1993. Importance of fungi in the diet of Gammarus pulex and Asellus aquaticus. Oecologia 96, 304–309.
Green, D.W., Williams, K.A., Pascoe, D., 1986. The acute and chronic toxicity of cadmium to different life history stages of the freshwater crustaceanAsellus aquaticus (L). Archives of Environmental Contamination and Toxicology 15, 465–471.
Ham, L., Quinn, R., Pascoe, D., 1995. Effects of cadmium on the predator-prey interaction between the turbellarian Dendrocoelum lacteum (Müller, 1774) and the isopod crustacean Asellus aquaticus (L.). Archives of Environmental Contamination and Toxicology 29, 358–365.
Hare, L., 1992. Aquatic Insects and Trace Metals: Bioavailability, Bioaccumulation, and Toxicity. Critical Reviews in Toxicology 22, 327–369.
Hargeby, A., 1990. Effects of pH, humic substances and animal interactions on survival and physiological status of Asellus aquaticus L. and Gammarus pulex (L.). Oecologia 82, 348–354.
Hargeby, A., Stoltz, J., Johansson, J., 2005. Locally differentiated cryptic pigmentation in the freshwater isopod Asellus aquaticus. Journal of evolutionary biology 18, 713–721.
Hart, B.T., Davies, S.H., 1981. Trace metal speciation in the freshwater and estuarine regions of the Yarra River, Victoria. Estuarine, coastal and shelf science 12, 353–374.
Hasu, T., Tellervo Valtonen, E., Jokela, J., 2006. Costs of parasite resistance for female survival and parental care in a freshwater isopod. Oikos 114, 322–328.
Hatje, V., Payne, T.E., Hill, D.M., McOrist, G., Birch, G.F., Szymczak, R., 2003. Kinetics of trace element uptake and release by particles in estuarine waters: effects of pH, salinity, and particle loading. Environment international 29, 619–629.
Haus, N., Zimmermann, S., Wiegand, J., Sures, B., 2007. Occurrence of platinum and additional traffic related heavy metals in sediments and biota. Chemosphere 66, 619–629. https://doi.org/10.1016/j.chemosphere.2006.07.097
Holdich, D.M., Tolba, M.R., 1981. The effect of temperature and water quality on the in vitro development and survival of Asellus aquaticus (Crustacea: Isopoda) eggs. Hydrobiologia 78, 227–236.
Hunting, E.R., Mulder, C., Kraak, M.H.S., Breure, A.M., Admiraal, W., 2013. Effects of copper on invertebrate–sediment interactions. Environmental pollution 180, 131–135.
Ieromina, O., Peijnenburg, W., De Snoo, G.R., Vijver, M.G., 2014. Population responses of Daphnia magna, Chydorus sphaericus and Asellus aquaticus in pesticide contaminated ditches around bulb fields. Environmental pollution 192, 196–203.
Joachim, S., Roussel, H., Bonzom, J.-M., Thybaud, E., Mebane, C.A., Van den Brink, P., Gauthier, L., 2017. A long-term copper exposure in a freshwater ecosystem using lotic mesocosms: Invertebrate community responses. Environmental toxicology and chemistry 36, 2698–2714.

Johnson, R.K., Wiederholm, T., Rosenberg, D.M., 1993. Freshwater biomonitoring using individual organisms, populations, and species assemblages of benthic macroinvertebrates. Freshwater biomonitoring and benthic macroinvertebrates, 40-158.

Kar, D., Sur, P., Mandai, S.K., Saha, T., Kole, R.K., 2008. Assessment of heavy metal pollution in surface water. International Journal of Environmental Science & Technology 5, 119–124.

Kaya, H., Akbulut, M., Selvi, K., Ileri, B., Duysak, M., 2014. Heavy Metal Accumulation, Biomarker Responses and Sensitivity to Oxidative Stress in Isopoda Asellus aquaticus from Saricay Creek (Canakkale-Turkey). Ekoloji Dergisi 23.

Ketelaars, H.A., Frantzen, N.M., 1995. One decade of benthic macroinvertebrate biomonitoring in the River Meuse. Netherland Journal of Aquatic Ecology, 29, 121-133.

Kiffney, P.M., Clements, W.H., 1996. Size-dependent response of macroinvertebrates to metals in experimental streams. Environmental Toxicology and Chemistry 15, 1352–1356.

Klaassen, C.D., Liu, J., Choudhuri, S., 1999. Metallothionein: an intracellular protein to protect against cadmium toxicity. Annual review of pharmacology and toxicology 39, 267–294.

Köhler, H.-R., 2002. Localization of metals in cells of saprophagous soil arthropods (Isopoda, Diplopoda, Collembola). Microscopy research and technique 56, 393–401.

Kolaříková, K., Stuchlík, E., Liška, M., Horecký, J., Tátosová, J., Hardekopf, D., Lapšanská, N., Hořická, Z., Hovorka, J., Mihaljevič, M., Fuksa, J.K., von Tümpling, W., 2012. Long-Term Changes in the Bioaccumulation of As, Cd, Pb, and Hg in Macroinvertebrates from the Elbe River (Czech Republic). Water Air Soil Pollut 223, 3511–3526.

Kuppusamy, M.R., Giridhar, V.V., 2006. Factor analysis of water quality characteristics including trace metal speciation in the coastal environmental system of Chennai Ennore. Environment international 32, 174–179.

Lagerspetz, K.Y., 2003. Thermal acclimation without heat shock, and motor responses to a sudden temperature change in Asellus aquaticus. Journal of thermal biology 28, 421–427.

Lagerspetz, K., Mattila, M., 1961. Salinity reactions of some fresh-and brackish-water crustaceans. The Biological Bulletin, 120, 44-53.

Lamas, S., Fernández, J.A., Aboal, J.R., Carballeira, A., 2007. Testing the use of juvenile Salmo trutta L. as biomonitors of heavy metal pollution in freshwater. Chemosphere 67, 221–228.

Lockwood, A.P.M., 1959. The osmotic and ionic regulation of Asellus aquaticus (L.). Journal of Experimental Biology, 36, 546-555.

Lukančič, S., Žibrat, U., Mezek, T., Jerebic, A., Simčič, T., Brancelj, A., 2010. Effects of exposing two non-target crustacean species, Asellus aquaticus L., and Gammarus fossarum Koch., to atrazine and imidacloprid. Bulletin of environmental contamination and toxicology 84, 85.

MacNeil, C., Dick, J.T.., Bigsby, E., Elwood, R.W., Ian Montgomery, W., Gibbins, C.N., Kelly, D.W., 2002. The validity of the Gammarus:Asellus ratio as an index of organic pollution: abiotic and biotic influences. Water Research 36, 75–84.

Maltby, L., 1995. Sensitivity of the crustaceans Gammarus pulex (L.) and Asellus aquaticus (L.) to short-term exposure to hypoxia and unionized ammonia: Observations and possible mechanisms. Water Research 29, 781–787.

Maltby, L., 1991. Pollution as a Probe of Life-History Adaptation in Asellus aquaticus (Isopoda). Oikos 61, 11.Mantoura, R.F.C., Dickson, A., Riley, J.P., 1978. The complexation of metals with humic materials in natural waters. Estuarine and Coastal Marine Science 6, 387–408.

Markert, B., Wappelhorst, O., Weckert, V., Herpin, U., Siewers, U., Friese, K., Breulmann, G., 1999. The use of bioindicators for monitoring the heavy-metal status of the environment. Journal of Radioanalytical and Nuclear Chemistry 240, 425–429.

Martin, T.R., Holdich, D.M., 1986. The acute lethal toxicity of heavy metals to peracarid crustaceans (with particular reference to fresh-water asellids and gammarids). Water Research 20, 1137–1147.

Meylan, S., Behra, R., Sigg, L., 2004. Influence of metal speciation in natural freshwater on bioaccumulation of copper and zinc in periphyton: a microcosm study. Environmental science & technology 38, 3104–3111.

Meylan, S., Behra, R., Sigg, L., 2003. Accumulation of copper and zinc in periphyton in response to dynamic variations of metal speciation in freshwater. Environmental science & technology 37, 5204–5212.

Migliore, L., de Nicola Giudici, M., 1990. Toxicity of heavy metals to Asellus aquaticus (L.)(Crustacea, Isopoda). Hydrobiologia 203, 155–164.

Moldovan, M., Rauch, S., Gómez, M., Antonia Palacios, M., Morrison, G.M., 2001. Bioaccumulation of palladium, platinum and rhodium from urban particulates and sediments by the freshwater isopod Asellus aquaticus. Water Research 35, 4175–4183.

Montañés, J.C., Van Hattum, B., Deneer, J., 1995. Bioconcentration of chlorpyrifos by the freshwater isopod Asellus aquaticus (L.) in outdoor experimental ditches. Environmental Pollution 88, 137–146.

Moore, J.W., 1975. The Role of Algae in the Diet of Asellus aquaticus L. and Gammarus pulex L. Journal of Animal Ecology 44, 719–730.

Naylor, C., Pindar, L., Calow, P., 1990. Inter-and intraspecific variation in sensitivity to toxins; the effects of acidity and zinc on the freshwater crustaceans Asellus aquaticus (L.) and Gammarus pulex (L.). Water research 24, 757–762.

Økland, K.A., 1978. Life history and growth of Asellus aquaticus (L.) in relation to environment in a eutrophic lake in Norway. Hydrobiologia 59, 243–259.

O'Neill, A.J., Galloway, T.S., Browne, M.A., Dissanayake, A., Depledge, M.H., 2004. Evaluation of toxicity in tributaries of the Mersey estuary using the isopod Asellus aquaticus (L.). Marine Environmental Research 58, 327–331.

Pagenkopf, G.K., 1983. Gill surface interaction model for trace-metal toxicity to fishes: role of complexation, pH, and water hardness. Environmental Science & Technology 17, 342–347.

Pagnanelli, F., Esposito, A., Toro, L., Veglio, F., 2003. Metal speciation and pH effect on Pb, Cu, Zn and Cd biosorption onto Sphaerotilus natans: Langmuir-type empirical model. Water research 37, 627–633.

Palma, P., Ledo, L., Alvarenga, P., 2015. Assessment of trace element pollution and its environmental risk to freshwater sediments influenced by anthropogenic contributions: the case study of Alqueva reservoir (Guadiana Basin). Catena 128, 174–184.

Pascoe, D., Carroll, K., 2004. Comparison of cadmium toxicity to Asellus aquaticus (L.) populations following 17 years isolation in pond and laboratory cultures. Bulletin of environmental contamination and toxicology 73, 167–173.

Peeters, E.T., De Jager, T.J., Beijer, J.A., Koelmans, A.A., 2000. Effects of benzo (a) pyrene and size of organic matter particles on bioaccumulation and growth of Asellus aquaticus. Archives of environmental contamination and toxicology 39, 307–314.

Persson, L., 1983. Food consumption and competition between age classes in a perch Perca fluviatilis population in a shallow eutrophic lake. Oikos 197–207.

Plahuta, M., Tišler, T., Pintar, A., Toman, M.J., 2015. Adverse effects of bisphenol A on water louse (Asellus aquaticus). Ecotoxicology and Environmental Safety 117, 81–88.Plahuta, M., Tišler, T., Toman, M.J., Pintar, A., 2017. Toxic and endocrine disrupting effects of wastewater treatment plant influents and effluents on a freshwater isopod Asellus aquaticus (Isopoda, Crustacea). Chemosphere 174, 342–353.

Qiao, P., Gobas, F., Farrell, A.P., 2000. Relative contributions of aqueous and dietary uptake of hydrophobic chemicals to the body burden in juvenile rainbow trout. Archives of environmental contamination and toxicology 39, 369–377.

Rainbow, P.S., 1995. Biomonitoring of heavy metal availability in the marine environment. Marine pollution bulletin, 31, 183-192.

Rask, M., Hiisivuori, C., 1985. The predation on Asellus aquaticus (L.) by perch, Perca fluviatilis (L.), in a small forest lake. Hydrobiologia 121, 27–33.
Rasmussen, J.J., Wiberg-Larsen, P., Baattrup-Pedersen, A., Cedergreen, N., McKnight, U.S., Kreuger, J., Jacobsen, D., Kristensen, E.A., Friberg, N., 2015. The legacy of pesticide pollution: an overlooked factor in current risk assessments of freshwater systems. Water research 84, 25–32.
Rauch, S., Morrison, G.M., 1999. Platinum uptake by the freshwater isopod Asellus Aquaticus in urban rivers. Science of The Total Environment 235, 261–268.
Resh, V.H., 2008. Which group is best? Attributes of different biological assemblages used in freshwater biomonitoring programs. Environmental Monitoring and Assessment 138, 131–138.
Richter, S., Nagel, R., 2007. Bioconcentration, biomagnification and metabolism of 14C-terbutryn and 14C-benzo [a] pyrene in Gammarus fossarum and Asellus aquaticus. Chemosphere 66, 603–610.
Roshchin, V.E., Mazelev, K.L., 1979. The influence of constant temperature on the embryonic growth of Asellus aquaticus L.(Crustacea)[Translation from: Vestsi Akademii Navuk Belorusskoi SSR, Seriya Biyal. 1979 (1) 128-130].
Rossi, L., Fano, A.E., 1979. Role of fungi in the trophic niche of the congeneric detritivorous Asellus aquaticus and A. coxalis (Isopoda). Oikos 380–385.
Ruchter, N., Zimmermann, S., Sures, B., 2015. Field Studies on PGE in Aquatic Ecosystems. Springer Berlin Heidelberg, pp. 351–360.
Sakan, S.M., Đorđević, D.S., Manojlović, D.D., Predrag, P.S., 2009. Assessment of heavy metal pollutants accumulation in the Tisza river sediments. Journal of environmental management 90, 3382–3390.
Sako, A., Lopes, L., Roychoudhury, A.N., 2009. Adsorption and surface complexation modeling of palladium, rhodium and platinum in surficial semi-arid soils and sediment. Applied Geochemistry 24, 86–95.
Santoro, A., Blo, G., Mastrolitti, S., Fagioli, F., 2009. Bioaccumulation of Heavy Metals by Aquatic Macroinvertebrates Along the Basento River in the South of Italy. Water, Air, and Soil Pollution 201, 19–31.
Savage, A., Hogarth, P.J., 1999. An analysis of temperature-induced fluctuating asymmetry in Asellus aquaticus (Linn.). Hydrobiologia 411, 139–143.
Simčič, T., Brancelj, A., 2006. Effects of pH on electron transport system (ETS) activity and oxygen consumption in Gammarus fossarum, Asellus aquaticus and Niphargus sphagnicolus. Freshwater Biology 51, 686–694.
Sket, B., 1994. Distribution of Asellus aquaticus (Crustacea: Isopoda: Asellidae) and its hypogean populations at different geographic scales, with a note on Proasellus istrianus. Hydrobiologia 287, 39–47.
Späth, R., Flemming, H.-C., Wuertz, S., 1998. Sorption properties of biofilms. Water Science and Technology 37, 207–210.
Steel, E.A., 1961. Some observations on the life history of Asellus aquaticus (L.) and Asellus meridianus Racovitza (Crustacea: Isopoda), in: Proceedings of the Zoological Society of London. Wiley Online Library, pp. 71–87.
Sures, B., Taraschewski, H., 1995. Cadmium concentrations in two adult acanthocephalans, Pomphorhynchus laevis andAcanthocephalus lucii, as compared with their fish hosts and cadmium and lead levels in larvae ofA. lucii as compared with their crustacean host. Parasitology Research 81, 494–497.
Sworobowicz, L., Grabowski, M., Mamos, T., Burzyński, A., Kilikowska, A., Sell, J., Wysocka, A., 2015. Revisiting the phylogeography of Asellus aquaticus in Europe: insights into cryptic diversity and spatiotemporal diversification. Freshwater Biology 60, 1824–1840.

Talbot, V., Magee, R.J., 1978. Naturally-occurring heavy metal binding proteins in invertebrates. Archives of environmental contamination and toxicology 7, 73–81.
Tipping, E., Rey-Castro, C., Bryan, S.E., Hamilton-Taylor, J., 2002. Al (III) and Fe (III) binding by humic substances in freshwaters, and implications for trace metal speciation. Geochimica et cosmochimica acta 66, 3211–3224.
Tolba, M.R., Holdich, D.M., 1981. The effect of water quality on the size and fecundity of Asellus aquaticus (Crustacea: Isopoda). Aquatic Toxicology 1, 101–112.
Tuccillo, M.E., 2006. Size fractionation of metals in runoff from residential and highway storm sewers. Science of the Total Environment 355, 288–300.
Tulonen, T., Pihlström, M., Arvola, L., Rask, M., 2006. Concentrations of heavy metals in food web components of small, boreal lakes. Boreal Environment Research 11, 185–194.
Turk, S., Sket, B., Sarbu, Ş., 1996. Comparison between some epigean and hypogean populations of Asellus aquaticus (Crustacea: Isopoda: Asellidae). Hydrobiologia 337, 161–170.
Turk-Prevorčnik, S., Blejec, A., 1998. Asellus aquaticus infernus, new subspecies (Isopoda: Asellota: Asellidae), from Romanian hypogean waters. Journal of Crustacean Biology 18, 763–773.
Usseglio-Polatera, P., Bournaud, M., Richoux, P., Tachet, H., 2000. Biomonitoring through biological traits of benthic macroinvertebrates: how to use species trait databases? Assessing the Ecological Integrity of Running Waters, 153-162. Springer, Dordrecht.
Van Ginneken, M., Blust, R., Bervoets, L., 2018. Combined effects of metal mixtures and predator stress on the freshwater isopod Asellus aquaticus. Aquatic Toxicology 200, 148–157.
Van Ginneken, M., Blust, R., Bervoets, L., 2017. How lethal concentration changes over time: Toxicity of cadmium, copper, and lead to the freshwater isopod Asellus aquaticus. Environmental Toxicology and Chemistry 36, 2849–2854.
Van Ginneken, M., De Jonge, M., Bervoets, L., Blust, R., 2015. Uptake and toxicity of Cd, Cu and Pb mixtures in the isopod Asellus aquaticus from waterborne exposure. Science of The Total Environment 537, 170–179.
Van Hattum, B., Van Straalen, N.M., Govers, H.A.J., 1996. Trace metals in populations of freshwater isopods: influence of biotic and abiotic variables. Archives of environmental contamination and toxicology 31, 303–318.
Veglio, F., Beolchini, F., 1997. Removal of metals by biosorption: a review. Hydrometallurgy 44, 301–316.
Verovnik, R., Sket, B., Trontelj, P., 2005. The colonization of Europe by the freshwater crustacean Asellus aquaticus (Crustacea: Isopoda) proceeded from ancient refugia and was directed by habitat connectivity. Molecular Ecology 14, 4355–4369.
Verrell, P.A., 1985. Predation and the evolution of precopula in the isopod Asellus aquaticus. Behaviour 198–202.
Vijver, M.G., Elliott, E.G., Peijnenburg, W.J., De Snoo, G.R., 2011. Response predictions for organisms water-exposed to metal mixtures: A meta-analysis. Environmental Toxicology and Chemistry 30, 1482–1487.
Warren, L.A., Haack, E.A., 2001. Biogeochemical controls on metal behaviour in freshwater environments. Earth-Science Reviews 54, 261–320.
Weltje, L., Oehlmann, J., 2006. Effects of endocrine disrupting compounds and temperature on the moulting frequency of the freshwater isopod Asellus aquaticus L.(Isopoda: Asellota). Acta Biologica Benrodis 13, 105–115.
Whitehurst, I., 1991. TheGammarus: Asellus ratio as an index of organic pollution. Water Research 25, 333–339.
Williams, W.D., 1962. Notes on the ecological similarities of Asellus aquaticus (L.) and A. meridianus Rac.(Crust., Isopoda). Hydrobiologia 20, 1–30.

Wolff, W.J., 1973. The distribution of Asellus aquaticus (L.) and Proasellus meridianus (Rac.) in the southwestern part of the Netherlands. Hydrobiologia, 42, 381-392.