Stoichiometric Ecotoxicology for a Multisubstance World

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Abstract **:**

Nutritional and contaminant stressors influence organismal physiology, trophic interactions, community structure, and ecosystem-level processes; however, the interactions between toxicity and elemental imbalance in food resources have been examined in only a few ecotoxicity studies. Integrating well-developed ecological theories that cross all levels of biological organization can enhance our understanding of ecotoxicology. In the present article, we underline the opportunity to couple concepts and approaches used in the theory of ecological stoichiometry (ES) to ask ecotoxicological questions and introduce stoichiometric ecotoxicology, a subfield in ecology that examines how contaminant stress, nutrient supply, and elemental constraints interact throughout all levels of biological organization. This conceptual framework unifying ecotoxicology with ES offers potential for both empirical and theoretical studies to deepen our mechanistic understanding of the adverse outcomes of chemicals across ecological scales and improve the predictive powers of ecotoxicology.

Keywords : nutrient ratios, elemental imbalances, toxicity, ecotoxicological models, multiple stressors

1 Introduction

2   Ecotoxicology is the study of the harmful effects of chemical contaminants on

3   individual organisms, populations, communities, and ecosystem processes (Cairns Jr

4   1988; Baird et al. 1996; Calow 1996; Relyea and Hoverman 2006; Gessner and Tlili

5   2016; Walker, Sibly, and Peakall 2016). Ecotoxicologists have found that toxicity may be

6   influenced by environmental factors such as ambient physico-chemical conditions

7   influencing contaminant bioavailability, organismal traits (e.g., differential

8   metabolism/sensitivity, trophic guild), and/or food webs or ecosystem structure (Newman

9   and Clements 2007). While these findings are often incorporated into regulatory actions,

10   many 'big-picture' ecological theories that cross all levels of biological organization have

11   not been consistently integrated into the field of ecotoxicology and subsequent

12   applications during chemicals assessment and management.

13   How organisms obtain the nutrition and energy needed for maintenance, growth,

14   and reproduction is a central concept in many ecological studies that examine organismal

15   physiology (Wagner et al. 2013), food webs (Phillips and Eldridge 2006), biodiversity

16   (Worm et al. 2002), and biogeochemical cycles (Sterner and Elser 2002). Given how

17   nutrition can be an important driver in many ecological studies, it is no surprise that

18   multiple different frameworks are used to examine how diets affect organismal

19   phenotypes. All nutritional ecological frameworks in the broadest sense examine how

20   nutrition affects the physiology, behavior, life-history, and eco-evolutionary responses of

21   organisms and their interactions with the environment (Raubenheimer, Simpson, and

22   Mayntz 2009). Frameworks such as optimal foraging and nutritional geometry have a

23   behavioral ecology foundation that focuses on optimizing fitness traits based on food

selection. Initially, nutritional geometry focused on controlled laboratory experiments to examine the optimal nutrition of two macronutrients (i.e., proteins, carbohydrates). More recent techniques have including adding a third nutrient to investigate more complex diets using the right-angle triangle (Raubenheimer 2011); however, much of the work has focused on organisms and is not readily scaled to communities and ecosystems. Other frameworks such as ecological stoichiometry (ES) that tracks elements and have a foundational focus on ecosystem science (Sterner and Elser 2002; Sperfeld et al. 2017). Early work in this area focused on interspecific competition and biogeochemical cycles (e.g., Elser et al. 1998) whereas more recent work includes a focus on the physiological changes that occur due to element-limited diets (Wagner et al. 2013). While ES is limited partly because some types of micronutrient limitation (i.e., essential fatty acids/amino acids) are not easily identified by studying elements alone, recent work has extended the ES modeling framework to incorporate concepts from nutritional geometry and has shown that macronutrients can be used as currencies (Anderson et al. 2020). Overall, the relative ease of tracking elements through food webs that can span all spatial levels of organization from genes to ecosystems (Hessen et al. 2013) makes ES an ideal framework to examine how toxicants and elemental stress affect organisms and ecosystems.

At the most fundamental level, ES is the study of imbalances between the elemental composition of available resources and what is required for organismal metabolism (Sterner and Elser 2002). In essence, imbalances between multiple available elements and organismal requirements of those elements strongly affect the physiology of many taxa and result in widely variable growth, reproduction, and survival (Frost et al.

2005; Wagner et al. 2013; Sterner and Elser 2002). Stoichiometric effects on organisms in turn alter ecosystems and biogeochemical cycles (Elser et al. 1998). If the organism elemental requirements are imbalanced with the elemental supply ratios, it would alter the nutrition of the organism and effect their excretion elemental ratios. For example, an imbalanced nitrogen:phosphorus (N:P) supply ratios can alter the nutrition of zooplankton consumers and their N and P excretion ratios (Elser and Urabe 1999). ES examines the causes and consequences of elemental imbalances across biological levels by linking cellular processes and organismal metabolism to population dynamics, predator-prey interactions, and ecosystem function (Sterner and Elser 2002).

As elemental ratios in resources vary widely and elemental limitation is widespread in nature, ecotoxicology may benefit from the incorporation of a stoichiometric perspective. Traditionally, toxicity experiments use nutrient-replete conditions, with the intention to eliminate nutrient limitation and elemental imbalances, because of the inherent assumption that nutrient limitation would interact with toxins or toxicants to alter results. While nutrient replete conditions may not be commonplace in natural ecosystems, only a few ecotoxicology studies have examined toxicity-elemental imbalance interactions. When explicitly included in single-species toxicity testing, the effect of the contaminant has been found to vary when there are elemental imbalances between the test organism and its nutritional resources (e.g., Hansen et al. 2008; Fulton et al. 2009; Fulton et al. 2010; Lessard and Frost 2012; Bian et al. 2013; Conine and Frost 2017). While this handful of examples demonstrate the possible uses of ES to improve our understanding of ecotoxicology, this past work represents only a small part of what could be a larger conceptually united subdiscipline: stoichiometric ecotoxicology. We

1 define stoichiometric ecotoxicology as a subfield in ecology that examines how

2 imbalances between organisms and their food resources interact with contaminant stress

3 in all levels of biological organization. Further, we define nutrients in this framework as

4 elements that are conserved across all spatial scales from genes to ecosystems.

5   One area of stoichiometric ecotoxicology that could be especially useful but

6 remains undeveloped is the application of formal mathematical approaches that couple

7 mass balance constraints to ecological processes. Stoichiometric models track multiple

8 elements to understand how elemental imbalances affect processes from cells to whole

9 ecosystems across multiple levels of biological organization. For example, at the

10 organism scale, mass-balance models couple information on physiological processes of

11 multiple elements (ingestion and assimilation) to determine animal elemental demands

12 and the optimal nutritional mixture needed for maximal growth (Sterner 1997; Frost and

13 Elser 2002; Frost et al. 2006). In addition, organismal processes are incorporated in

14 dynamical models that quantitatively link energy flow and elemental cycling to predator-

15 prey interactions (Loladze, Kuang, and Elser 2000; Elser et al. 2012; Muller et al. 2001).

16 These models could be extended to examine the ecological effects of natural or

17 anthropogenic contaminants by modifying parameters relative to chemical concentration

18 or body burden. Similarly, classical ecotoxicological models can be expanded to

19 incorporate stoichiometric constraints by modifying parameters related to nutrient

20 limitations. Integrative modeling approaches would allow for examination of interactive

21 effects of nutrients and contaminants on organismal physiological state, competitive

22 interactions, and whole-ecosystem mass balance. Additionally, these models may help

23 determine critical contaminant concentration thresholds that regulate biomass, growth

1 rates, ecosystem function, and the biological transfer of toxins through ecosystems

2 (Andersen, Elser, and Hessen 2004; Evans-White et al. 2009; Suzuki-Ohno, Kawata, and

3 Urabe 2012). Current models are beginning to incorporate stoichiometric constraints to

4 understand the ecological effects of chemical toxicity ((Peace, Poteat, and Wang 2016;

5 Hassan et al. 2018), as discussed below) and this effort calls for conceptual, theoretical,

6 and empirical development.

7   Here we examine opportunities to develop stoichiometric ecotoxicology by

8 coupling concepts and approaches commonly used in ES to ask ecotoxicology questions.

9 In particular, we present a conceptual overview of the links between elemental

10 stoichiometry and toxicology, discuss recent advances in stoichiometric ecotoxicology

11 research, and explore possible areas of future development, including the development

12 and application of new or modified mathematical models. Altogether we show the need

13 and promise of stoichiometric ecotoxicology to provide the impetus for its future

14 development.

15
16 **The conceptualization of stoichiometric ecotoxicology**

17   While the potential value of examining interactions between stoichiometry and

18 toxicology has been noted (see above), a next step is to further develop the empirical

19 basis of stoichiometric ecotoxicology through the study of a diverse range of organisms, a

20 broader range of toxic contaminants, and with more types of elemental imbalances. The

21 underlying rationale and unifying concept of this research field is that the supply of

22 multiple resources can alter or be altered by the effects of chemical toxicity on

23 organisms, populations, communities, and ecosystems (Figure 1). As further development

24 of this field will require examining these connections using a combination of empirical

1 and theoretical approaches, a starting point is to identify the possible types and nature of

2 interactions between toxins and toxicants and unbalanced elemental supply. In addition,

3 this perspective is not taxa- or trophic-level specific, as we envision it being applied to

4 toxicological research on all organisms including producers, consumers, detritivores, and

5 predators. Here we provide an initial overview of stoichiometric ecotoxicology and its

6 potential applications across the various levels of biological organization (Figure 1).

7 Also, we have compiled a list of studies that implicitly or explicitly investigate the role of

8 stoichiometric constraints on the response of organisms to diverse toxic substances,

9 including pesticides, pharmaceuticals, essential and nonessential metals, and natural

10 substances such as plant secondary compounds (Table 1). These studies represent a

11 starting point from which we identify the types of ecotoxicological questions that a

12 stoichiometric framework allows us to ask at multiple levels of biological organization.

13

14 **Organismal stoichiometry and ecotoxicology.** Organismal physiology and life-history

15 are strongly affected by both available food resources (Figure 1, arrow 2) and

16 contaminants (Figure 1, arrow 3), and these alterations to metabolism may interact to

17 create more variation in toxicity. The nature of these interactive effects largely depends

18 on the nutrients under consideration, the organismal traits, and the nature of the

19 contaminant. For example, similar to the concentration addition mechanism of

20 contaminant mixtures (Altenburger et al. 1996), if a toxicant and elemental stress affect

21 the same metabolic pathways it could lead to an additive or synergistic stress response.

22 Additionally, exposure to toxicants may activate detoxification pathways within an

23 organism that can involve the synthesis of compounds rich in carbon (C) or other

24 elements. If these detoxification compounds mainly involve C, they may help alleviate

1 elemental imbalances through disposing of excess C supplies that accompany elemental

2 imbalances. Alternatively, excessive C demands could eventually lead to energy

3 limitation and reduced nutrient use efficiency. If these detoxification requirements

4 involve a non-C element in short supply, toxicity could exacerbate elemental limitation or

5 impair repair in exposed organisms.

6   The presence of contaminants may also directly alter the nutritional quality of

7 food resources available to both primary producers and consumers (Figure 1, arrow 1).

8 Numerous metals produce non-soluble complexes with elements that are important

9 nutritionally, such as P. For example, aluminum and phosphate form non-soluble

10 complexes, and so the presence of aluminum may reduce P supply and lead to skewed

11 N:P ratios available to primary producers. This could result in increased P- limitation of

12 organismal growth and altered ecological processes (Kopácek et al. 2000; Clivot et al.

13 2014). Indeed, aluminum additions have been used to intentionally reduce P availability

14 in lakes for some time, despite its known toxicity (e.g., Steinman and Ogdahl 2008).

15   Contaminants may also directly alter the C:N:P ratios of producers and consumers

16 by altering their physiology (e.g., via increased maintenance costs associated with

17 recovery from toxin exposure; Figure 1 arrow 3). Das et al. (2014) found reduced C:P

18 and N:P content of phytoplankton communities exposed to silver nanoparticles, while

19 Cherchi et al. (2015) found opposite trends in the cyanobacteria, *Anabaena variabilis,*

20 exposed to nano-titanium dioxide. Fulton et al. (2010) working on the aquatic

21 macrophyte, *Lemna gibba*, showed that triclosan (a common antimicrobial agent) caused

22 changes in tissue C:P ratios, depending on the N:P ratio of the culture medium. Exposure

23 to ionic silver has also been shown to reduce leaf litter microbial conditioning, resulting

1   in higher leaf litter C:N and C:P ratio (Arce-Funck et al. 2013). Nutrient stoichiometry

2   has also been shown to alter the toxicity of contaminants to plants (Fulton et al. 2009;

3   Fulton et al. 2010; Mkandawire and Dudel 2012; Bian et al. 2013; Brooks, Fulton, and

4   Hanson 2015), especially through changes in growth rate (Brooks, Fulton, and Hanson

5   2015). Finally, even consumers can exhibit altered stoichiometry when exposed to

6   contaminants as seen in the larvae of the damselfly *Enallagma cyathigerum* (i.e., a food

7   resource for predators), which had higher N:P ratios when exposed to the pesticide

8   chlorpyrifos (Janssens, Op de Beeck, and Stoks 2016).

9   Elemental resource supply affects the production of secondary toxic metabolites

10   in plants and phytoplankton, which is further altered in the presence of anthropogenic

11   contaminant exposure. Several studies have investigated the stoichiometry of plant

12   secondary compounds, such as polyphenols (Bryant, Chapin III, and Klein 1983; Rivas-

13   Ubach et al. 2012; Nuttens et al. 2016). For example, Nuttens et al. (2016) showed that

14   higher N:P ratios in culture media led to lower concentrations of phenolic compounds in

15   the aquatic plant, *Myriophyllum spicatum*, and that this effect disappeared when plants

16   were exposed to the herbicide mesosulfuron. Elemental stoichiometry is also associated

17   with eukaryotic and prokaryotic toxin production (Van de Waal et al. (2009; 2014). As

18   many of these toxins are N-rich (i.e. low C:N), the concentration and supply ratios of

19   C:N, and N:P influence the production of these toxins, with high N supply compared to

20   demand increases N-rich toxin production (Van de Waal et al. (2009; 2014). In turn,

21   toxin production is sensitive to anthropogenic exposure. For example, *Microcystis*

22   populations produce more of the cyanotoxin, microcystin-LR, when exposed to the

23   antibiotic amoxicillin (Liu et al. 2014). These studies highlight the complex interactions

1 between elemental supply ratios, secondary metabolic toxin production physiology, and

2 their interaction with anthropogenic contaminant exposure.

3   Ecotoxicological investigations are often divided into two broad categories,

4 studies of contaminant exposure (bioconcentration, bioaccumulation, biomagnification)

5 and of contaminant effects (nonlethal and lethal), each of which can be influenced by

6 stoichiometric constraints. The effects of elemental imbalances on metabolism could also

7 alter the uptake (Figure 1, arrow 3), biotransformation, and elimination of contaminants

8 (Figure 1, arrow 4) with implications for contaminant fate and toxicity. For example, the

9 uptake of silver nanoparticles by algae, through an unknown mechanism, was higher

10 under P-rich compared to P-poor conditions (Conine and Frost 2017). In this case, P-rich

11 algae significantly reduced the toxicant concentration in water that, in turn, altered the

12 toxic impacts on animal consumers. Contaminants may also alter the uptake of elements

13 through modification of feeding behavior or digestive and assimilative processes (e.g.,

14 Frost and Tuchman 2005; Fulton et al. 2010). Several examples of this have been

15 explored for *Daphnia*, which alters its feeding rates in response to P availability

16 (Darchambeau, Faerøvig, and Hessen 2003; Karimi et al. 2007) and therefore may

17 experience increased exposure to food-borne toxicants (Karimi et al. 2007).

18 Stoichiometric constraints might also have indirect impacts on the bioaccumulation of

19 contaminants by organisms. The bioaccumulation/bioconcentration of lipophilic toxicants

20 can be affected by N or P-limitation that cause the accumulation of lipid stores both

21 producers (Kilham 1998) and consumers (Wagner et al. 2014). Indirect effects have been

22 investigated using both theoretical models (Peace, Poteat, and Wang 2016) and

23 experimental approaches (Wang and Dei 2006; Karimi et al. 2007; Arce-Funck et al.

1   2018). Those studies concluded that well-balanced elemental resources increase

2   organismal growth and dilute contaminants due to increased biomass.

3   Food quality and contaminants interact to affect the survival, growth, and

4   reproduction of producers and consumers. For example, *Daphnia* feeding on a low P diet

5   had a higher LC50 (lethal concentration required to kill 50% of the population) compared

6   to those feeding on P-rich diets when exposed to fluoxetine (Hansen et al. (2008). While

7   it is unknown what mechanism provides this decrease in toxicity, it has also been

8   displayed with *Daphnia* and other toxicants (e.g., glyphosate; (Lessard and Frost 2012)).

9   However, elemental limitation does not always confer protection against toxicity in

10   *Daphnia.* For example, elemental limitation increases toxicity when *Daphnia* were

11   exposed to imidacloprid (Ieromina et al. 2014) and silver nanoparticles (Conine and Frost

12   2017). This food quality contaminant interaction is not specific to planktonic consumers

13   and has also been found in a detritivore invertebrate, *Gammarus fossarum*. A higher

14   tolerance occurred in *G. fossarum* consuming high P diets after an acute exposure to

15   dissolved silver (Arce-Funck et al. 2016). While feeding on high P-resources during a

16   chronic, environmentally realistic exposure to cadmium tended to synergistically increase

17   the impact of the contaminant (Arce-Funck et al. 2018).

18   These examples highlight the need for controlled studies that examine each of the

19   many factors (e.g., timing, nutrient, and contaminant concentration gradients, exposure

20   duration) that may be important in understanding toxicity in a stoichiometric context.

21   However, these examples also suggest that organismal responses to both stoichiometric

22   and toxic stressors largely depend on the nature of the toxic compounds investigated,

23   duration of exposure, and the organism's biology. Coupled to these controlled studies, we

1   see the importance of future studies aimed at understanding environmental gradients of

2   chemical contaminants and complexity, particularly those substances with biological

3   activities mechanistically capable of altering molecular targets (e.g., transporters, efflux

4   pumps) and ecological functions in aquatic ecosystems (Rosi-Marshall and Royer 2012).

5   Similarly, future studies should consider environmental gradients of elements, including

6   conditions of elemental limitation and excess, that can alter growth rates, growth

7   efficiencies, and the ability of organisms to withstand changes in maintenance

8   requirements and other toxicant-induced stressors.

9

10   **Ecotoxicology and the stoichiometry of populations and communities.** Elemental

11   imbalances on producers and consumers have strong effects on their population dynamics

12   by altering population growth rates, maximum population size, and population

13   permanency. Such stoichiometric effects could modify adverse outcomes elicited by

14   contaminants at the population and community levels (Figure 1, arrow 6). For example,

15   populations of P-limited green algae are more sensitive to ibuprofen and less sensitive to

16   common high blood pressure and epilepsy medications compared to P-replete algae

17   (Grzesiuk, Wacker, and Spijkerman 2016). These indirect, elemental-driven differences

18   in toxicity could alter biomass production in lower trophic levels and food quality

19   available for predators, which add to the direct effects of the elements themselves.

20   Different species are also known to exhibit varying sensitivities to elemental

21   imbalances and/or contaminants, which will alter species composition in food webs and

22   community-level interactions (Figure 1, arrow 11). For example, antibiotic exposure of a

23   microbial community found on decomposing leaves resulted in a community shift in

1 favor of fungi over bacteria (Bundschuh et al. 2009). The antibiotic-exposed leaves with

2 more fungi were then preferentially consumed by invertebrate shredders, which suggests

3 interactive effects of elements and toxicants on the population and community dynamics

4 of heterotrophic food webs (Bundschuh et al. 2009). Changes in population size either

5 from poor nutrition associated with elemental imbalances or due to chemical toxicity may

6 also affect the total uptake and sequestration of contaminants with subsequent effects on

7 the exposure of other organisms (Figure 1, arrow 8). Despite their potential to occur,

8 community-level effects of stoichiometric toxicology are perhaps the least studied among

9 the different levels of biological organization.

10

11 **Ecosystems and stoichiometric ecotoxicology.** The field of ecological stoichiometry has

12 long studied how changes in the elemental nutrition of animals could affect the rate and

13 ratios of elemental cycling (Figure 1, arrow 5), which feedback into the supply of

14 elements available at the base of the food web (Figure 1, arrow 7; (Elser and Urabe

15 1999)). Similarly, toxic chemicals may alter metabolic states or the elemental

16 assimilation and excretion, with effects on the rate and ratios of elemental recycling

17 (Taylor et al. 2016). While this could be due to direct changes in organism metabolism, it

18 may also be mediated through changes to populations (Figure 1, arrow 9) or community

19 composition (Figure 1, arrow 10) as different life-stages and species may have distinct

20 stoichiometries (Bullejos et al. 2014).

21   The transfer of energy and matter across habitat boundaries, such as from aquatic

22 to terrestrial communities is well documented. Classic examples include aquatic insect

23 emergence being fed upon by riparian spiders (Kato et al 2003, Marczak and Richardson

1   2007) and birds (Murakami and Nakano 2002), or reciprocal transport from land to water

2   by terrestrial insects feeding trout (Nakano and Murakami 2001) and hippopotamus

3   transfer of savanna grassland nutrients to sub-Saharan African streams (Sabalusky et al.

4   2015). Given the breadth of knowledge linking nutrient availability to population and

5   community structure, as detailed above, it is apparent that C:N:P ratios of basal resources

6   likely influence organism and material movement across habitat boundaries.

7   Ecotoxicology can be linked to these subsidies as organically bound contaminants (e.g.,

8   PCBs, Walters et al. 2008, 2009) often accompany these movements of organisms. Yet,

9   to date, few studies have investigated questions related to these potential connections

10   (Arce-Funck et al., 2013; Fernandes et al., 2009) between biomass and composition of

11   organisms and toxins or toxicants, and the nutrient stoichiometry of food resources. There

12   is thus a need to better understand how stoichiometric mechanisms may alter the results

13   of ecotoxicological studies at the community and ecosystem levels.

14

Modeling Advances in Stoichiometric Ecotoxicology

16   Throughout its history, mathematical modeling has been a powerful tool with aims to

17   provide ways to organize ideas, develop hypotheses, and offer a qualitative understanding

18   of complex systems while making useful predictions. While traditional ecotoxicological

19   mechanistic modeling efforts take a variety of approaches to predict the effects of diverse

20   chemical contaminants on organismal responses (usually survival, growth, and

21   reproduction), many models assume, implicitly or explicitly, a nutrient-rich environment

22   where elemental limitation is absent. This assumption of a nutrient-replete environment

23   likely reflects the norm for empirical ecotoxicity studies, as noted above, where nutrient

1 media (in the case of plants, cyanobacteria, and algae) or food (for invertebrates and fish)

2 is sufficiently nutrient-rich to not overtly stress model organisms. In contrast, ES models

3 explicitly include elements to discover how the elemental composition of organisms

4 shapes their ecology, population dynamics, and ecosystem function. These models have

5 not generally considered contaminant effects on the causes and consequences of

6 elemental imbalances on organisms. Here we present brief overviews of models in these

7 two fields, consider recent modeling efforts that aim to integrate them, and discuss future

8 directions.

9

10 **Ecotoxicological Modeling.** The development of ecotoxicological models over the last

11 few decades has significantly contributed to interpreting how contaminants move and

12 accumulate throughout aquatic foods webs, as well as how they affect organisms (Hallam

13 and De Luna 1984; Mackay, Puig, and McCarty 1992; Gobas 1993; Ankley et al. 1995;

14 Wang, Fisher, and Luoma 1996; Kooijman and Bedaux 1996; Arnot and Gobas 2004;

15 Arnot and Gobas 2006; Pieters et al. 2006; Ashauer, Boxall, and Brown 2007; Wang et

16 al. 2008; Ashauer and Brown 2008; Bontje et al. 2009; Borgå et al. 2014; Huang, Wang,

17 and Lewis 2014, 2015; Huang and Wang 2016). These models take a variety of

18 approaches ranging from steady-state models of simplified systems with one to two

19 ordinary differential equations to more complex size-structured systems of partial

20 differential equations. While some of these models focus solely on contaminant exposure

21 (i.e. Arnot and Gobas 2006 balances uptake, assimilation, and elimination processes to

22 model bioaccumulation and Wang, Fisher, and Luoma 1996 uses kinetic models to

23 predict the bioaccumulation of trace elementals in mussels), many models simultaneously

model both contaminant exposure and effects (i.e. Huang, Wang, and Lewis 2015

dynamically model contaminant concentrations in organisms and how these body burdens

impact population dynamics). One of the best-tested modeling frameworks

systematically applied in ecotoxicology follows Dynamic Energy Budget (DEB) theory

and has been included in international risk-assessment guidance for its relevance in

analyzing ecotoxicity data (Jager, Gudmundsdottir, and Cedergreen 2014). While DEB

theory describes energy flows within an individual organism (Kooijman and Kooijman

2010; Nisbet et al. 2000), it aims to capture the quantitative aspects of metabolism and is

explicitly based on the principles of conservation of energy and mass (Sousa et al 2010).

The model can be applied and adapted to any organism with changes in species-specific

parameters. A recent Scientific Opinion for the European Food Safety Authority (EFSA)

noted the potential applicability of DEB models coupled to toxico-kinetic/toxicodynamic

effect models for regulatory risk assessment (Ockleford et al. 2018).

The "Add-my-pet" online database serves as a curated and constantly expanding

inventory of DEB parameters for thousands of species

(https://www.bio.vu.nl/thb/deb/deblab/add_my_pet/index.html, 2653 species, November

2020). The standard model describes how an organism ingests food, assimilating some of

the food as energy, and egesting the rest. Inside the organism, the assimilated energy is

stored in "reserve" components from which the organism allocates energy towards two

branches: growth and somatic maintenance and maturation/reproduction and maturity

maintenance. The independence of the reserve dynamics of food availability is a key

aspect of DEB theory and provides the individual with some protection against

environmental fluctuations and some control over its own metabolism (Sousa et al. 2010).

1   Contaminant effects are integrated into DEB models (nicknamed "DEBtox", see

2 Jager and Zimmer (2012) and Jager (2016)) either by allowing a toxicant to impact

3 (change the value of) DEB parameters (e.g. (Jager et al. 2014)) or through the addition of

4 sub-models that connect the DEB model to toxicant processes (Klanjscek et al. 2012;

5 2013). Moreover, DEBtox has been used to investigate the effects of multiple stressors at

6 population and ecosystem scales. When stressors (e.g. food and contaminant) interact,

7 they can have different effects (depending on the mode of action) at higher levels

8 (individual, population, or ecosystem) compared to the stressor in isolation, as was

9 demonstrated for *Daphnia* models by (Martin et al. 2014). It is unknown *a priori* if the

10 effects will be additive, synergistic, or antagonistic (Galic et al. 2018). In Galic et al.

11 (2017), for example, the exposure to toxicants targeting maintenance, feeding, and

12 reproduction on a freshwater amphipod (*Gammarus pseudolimnaeus*) disproportionately

13 weakened consumer-resource interactions under warming at the ecosystem level but had

14 no effect at the population level.

15   While these ecotoxicology modeling efforts have enhanced understandings and

16 improved predictions of the adverse effects of contaminants, they often neglect to

17 incorporate stoichiometric constraints. On the other hand, the following section describes

18 stoichiometric modeling approaches, which have largely been developed without

19 considering contaminant stressors.

20

21 **Ecological Stoichiometric Modeling**. An approach often used to study the effects of

22 elemental imbalances on plants, animals, and microbes involve tracking the intake,

23 acquisition, and release of multiple elements simultaneously. These models explicitly

1   incorporate a mass balance perspective insomuch that elements acquired in excess of

2   growth, reproduction, or maintenance will be released (Frost, Xenopoulos, and Larson

3   2004; Frost and Tuchman 2005). Following this, element-specific uptake or consumption

4   can be coupled to elemental use for new biomass or maintenance to yield estimates of an

5   ideal resource mixture that maximizes growth or reproduction (Sterner 1997; Frost and

6   Elser 2002; Anderson and Hessen 2005). By altering the elemental mixture available for

7   acquisition, these models yield predictions about the strength and direction of elemental

8   constraints on organismal performance including slower growth, reduced reproduction, or

9   alterations in nutrient release.

10   Elemental imbalances and their constraints across trophic levels have been

11   incorporated into dynamical system models of population as well. Andersen (1997) took

12   the classical Rosenzweig MacArthur variation of the Lotka-Volterra equations and

13   incorporated nutrient-deficient growth by modifying the density dependence of the

14   producer's growth rate and the grazer's growth efficiency. Following Andersen's

15   approach, Loladze, Kuang, and Elser (2000) formulated a producer-grazer model (LKE

16   model) of the first two trophic levels of an aquatic food chain (algae-*Daphnia*) that

17   incorporated the assumption that both producers and grazers are chemically

18   heterogeneous organisms composed of two essential elements, C and P. The model

19   allows the P:C of the producer to vary above a minimum value but restricted the P:C

20   variation in grazer biomass. Introduction of these stoichiometric constraints significantly

21   affects the population dynamics and stability properties of the system, such as exhibition

22   of the paradox of "energy enrichment" empirically supported by Urabe et al. (2002).

1   The growing library of models that have extended the LKE model under the

2 theory of ES is mechanistic and robust. These models have incorporated dynamical

3 consequences of elemental limitation (Loladze, Kuang, and Elser 2000) and effects of

4 elements in excess (Peace et al. 2013). They have also been expanded to incorporate

5 multiple elements (Grover 2002; 2003; 2004) and two competing species (Loladze et al.

6 2004; Xie, Fan, and Zhao 2010). They can explicitly track free elements in the

7 environment (Wang, Kuang, and Loladze 2008; Peace, Wang, and Kuang 2014), and

8 determine important trophic transfer efficiencies (Peace 2015). The ES modeling

9 framework has been expanded to include spatial dynamics (Rana et al. 2018), incorporate

10 multi-stage dynamic processes (Jiang et al. 2019), and capture evolutionary dynamics

11 (Yamamichi et al. 2015). These stoichiometric models incorporate the effects of both

12 food quantity and food quality into a single framework that produces rich dynamics (i.e.

13 stabilizing predator-prey systems) and allow more than one predator species to coexists

14 on a single prey, producing multiple basins of attraction and bi-stability, exhibiting

15 multiple types of bifurcations, and allowing evolutionary rescue. This ES modeling

16 framework applies equally to phenomena at sub-organismal levels as well as the whole

17 biosphere level (Sterner and Elser 2002). Despite these advances, models developed

18 under the ES framework typically disregard the effects of contaminants. The above

19 briefly summaries highlight the fact that the majority of modeling efforts in

20 ecotoxicology and ES have largely been developed separately. Below we discuss the

21 opportunities for theoretical approaches to better integrate these two fields within single

22 modeling frameworks.

23

1   **Stoichiometric Ecotoxicological Modeling**. While there are modeling efforts that

2   consider contaminant stressors along with element limitation (i.e. Bontje et al. (2009) and

3   Ankley et al. (1995)), these models are not explicitly stoichiometric and do not allow for

4   multiple elemental constraints where the element limiting growth can change with

5   environmental element and light availabilities. Here, we propose integrative modeling

6   frameworks with promising potential to explicitly incorporate elements and contaminants

7   as well as integrative effects between them.

8   Existing theoretical frameworks in ES and ecotoxicology actually have similar

9   structures but they utilize different perspectives. For example, ES uses an element

10   perspective to develop mass-balance models of threshold elemental ratios (i.e. Frost,

11   Xenopoulos, and Larson 2004, Frost and Tuchman 2005) and ecotoxicology uses a

12   contaminant perspective to develop mass-balance models of bioaccumulation (i.e. Gobas

13   1993). While the mass-balance modeling approaches here are similar, they employ

14   different assumptions on parameters as the biological regulation of elements can

15   significantly differ from regulation of contaminants. Both fields use dynamical

16   population models, where ES uses a nutrient perspective to track essential elements

17   throughout food webs and their influence of population dynamics (i.e. Loladze, Kuang,

18   and Elser 2000) and Ecotoxicology uses a contaminant perspective to track the fate of

19   contaminants throughout food webs and their effects on population dynamics (i.e. Huang,

20   Wang, and Lewis 2014, 2015). These modeling approaches can be unified by integrating

21   contaminants into the ES models and/or by integrating nutrients into the ecotoxicological

22   model as shown in Figure 2.

1   DEB theory offers a modeling framework with the potential to analyze

2   ecotoxicological issues in which stoichiometry plays an important role. This would

3   require a more detailed or explicit stoichiometric framework than is currently

4   incorporated in DEBtox studies. The standard DEB model is the simplest, non-

5   degenerated, model among a family of models (Marques et al. 2018) that describes the

6   full life cycle of an animal feeding on a single food source with constant elemental

7   composition. Thus, this model does not generally incorporate or examine stoichiometric

8   mechanisms per se. However, multiple-reserve (instead of single-reserve) DEB models

9   have been developed to consider stoichiometry explicitly. Kooijman (1998) proposed a

10   generalizable model of two reserves, specifically citing applications to element-limited

11   algal growth, such as the classic Droop models (Droop 1974). Sperfeld et al. (2017)

12   described a DEB model with separate nutrient reserves (C and N) coupled with metabolic

13   pathways that may be impacted by changes in C or N mass balance. DEB models that

14   consider stoichiometric constraints have been developed for a wide variety of

15   applications, ranging from tracing isotope flows (Pecquerie et al. 2010) to describing the

16   symbiosis between coral and photosynthetic algae (Muller et al. 2009; Cunning et al.

17   2017) to analyzing stoichiometric constraints on population dynamics (Kuijper et al.

18   2004a; 2004b; Kooijman 1998; Kooijman, Andersen, and Kooi 2004). However, to our

19   knowledge, no study has utilized a stoichiometrically-explicit DEB model to explain or

20   predict ecotoxicological responses or adverse outcomes.

21   ES offers a complementary modeling framework to investigate the influence of

22   elemental imbalances on responses of organisms to contaminants while simultaneously

23   considering the effects of contaminants on ecosystem processes (Danger and Maunoury-

Danger 2013). Toxicity could be incorporated into ES models by altering functions of

organismal nutrient intake and/or by changing element-metabolism relationships with

chemical exposure. For example, Frost and Tuchman (2005) used this type of model to

demonstrate how reduced growth and increased element release rates from two benthic

invertebrates resulted from lower assimilation efficiencies in animals consuming nutrient-

poor leaf material grown under elevated CO_2 concentrations. Peace, Poteat, and Wang

(2016) and Hassan et al. (2018) developed models that incorporated the effects of

concurrent element and toxicant stressors on population dynamics and the trophic transfer

of toxicants. These models dynamically model the bioaccumulation of toxicants under

varying nutrient resources, capturing stoichiometric contaminant exposure dynamics, and

incorporate the effects of toxicant and nutrient stressors into growth dynamics. In these

models, elemental changes in the environment impact organismal growth rates and thus

population dynamics, which can drive changes in contaminant concentrations within the

organisms. Additionally, they captured and explored a phenomenon called the Somatic

Growth Dilution (SGD) effect, observed empirically (Karimi et al. 2007), where

organisms experience a greater than proportional gain in biomass relative to toxicant

concentrations when consuming food with high nutritional content vs. low quality food.

In addition to influencing life-history traits, specific contaminants can impact organism

elemental composition, (Ni, Kelly, and Arhonditsis 2017; Baudrot et al. 2018; Danger

and Maunoury-Danger 2013). Future model iterations should consider the impact of

contaminants on organism elemental compositions. While there have been few

toxicological applications, ES models appear to have considerable potential to

1   significantly improve our understanding of the effects that chemical contaminants have

2   on organisms and ecosystems (Hansen et al. 2008).

3

4   **Areas of future research**

5   Organisms, populations, communities, and ecosystems all function in a world of

6   contaminant and nutrient mixtures. We are now at a point where we can aim for a more

7   mechanistic understanding of the combined effects of contaminants and elements at each

8   level of biological organization. To achieve meaningful advances, some of the

9   fundamental discoveries and models developed in the fields of ES and ecotoxicology

10 should be considered and, if possible, merged into a unified framework, as proposed in

11 Figure 2. These considerations could include: 1) determining nonlinear responses to

12 elements *and* contaminants and their interactions, 2) potential feedbacks between the

13 effects of elements and contaminants at different scales, and 3) integrating molecular

14 responses to better link to and understand the underlying associated biological changes.

15 The first steps would help define an integrated conceptual framework and inform specific

16 research goals to advance our understanding of the effects of contaminant-nutrient

17 combinations that more closely reflect natural environments.

18   Nonlinearities in the effects of individual elements and contaminants could alter

19 our predictions of interactions between these substances. At the organismal level,

20 nonlinear physiological responses to individual contaminants have been well documented

21 for decades. A classic example is the hormesis response in which organisms exhibit a

22 positive response to a contaminant at low doses and a negative, deleterious response at

23 higher levels (Calabrese and Baldwin 2002). More recent work suggests biological

adaptation as a potential mechanistic explanation for the otherwise phenomenological

hormetic response (Calabrese and Mattson 2017). Another landmark study examined and

defined nonmonotonic responses to a suite of endocrine disrupting contaminants

(Vandenberg et al. 2012). Classic models highlight nonlinearities in physiological

responses to essential and nonessential metals (Karimi and Folt 2006). Recent work also

suggests that nonlinearities are not limited to toxicants and can also occur in response to

variable element (e.g., P and N) regimes. Specifically, high inputs of N and P can cause

negative effects in forms that are not otherwise explained by other nutrient deficiencies

and that would not be expected based on ES theory (Costello et al. 2018). Additional

studies are needed to understand the negative effects of high element doses, particularly

as global element supplies continue to increase, and eutrophication of diverse ecosystems

become more evident (Howarth 2008). Overall, nonlinear responses are neither rare, nor

limited to certain types of substances, particularly when substances interact with multiple

biological targets, and therefore have high potential to influence the combined effects of

multiple nutrients and contaminants. Thus, there is a need to develop predictive models

that examine and explain the influence of nonlinear responses on the combined and

interactive effects of multiple substances.

Nonlinear interactions and dynamics illustrate how complexities in the behavior

and effects of elements and toxicants pose a challenge for how we distinguish among

different types of substances. In some cases, substances considered to be elements

function more like contaminants (Karimi and Folt 2006; Stumm and Morgan 2012).

Several different classification systems exist that attempt to make clear, useful

distinctions among types of substances based on different characteristics. For

convenience in this paper, we have used perhaps the most simplistic classification system

comprised only of nutrients versus contaminants. However, developing a unified

framework that combines knowledge from the ES and ecotoxicology fields requires us to

revisit common classification systems in terms of how they may help us understand the

behavior and effects of elements and toxicants in mixtures. This would help us organize

and predict potentially complex, higher-order interactions among substances. Part of this

could be mediated through a better understanding of the molecular components involved

in the processes underlying element and toxicant effects as described by the various DEB

processes. Well-known examples of such systems include: 1) classes of metals (e.g.,

Class A, B), 2) macro-elements, essential trace metals, nonessential trace metals (Karimi

and Folt 2006; Karimi 2010), 3) organic contaminant functions based on K_{ow} values, and

4) contaminant functions based on specific molecular interactions. Moving toward a

stoichiometric ecotoxicology framework requires us to define substances in ways that

help us understand whether there are emergent patterns in how different types of

substances interact and are organized at different scales.

A recent development in ecotoxicology to conceptualize "big-picture" impacts of

contaminants has been made through the adverse outcome pathway (AOP) framework

(Ankley et al. 2010). This framework could offer a way to classify substances in the

context of stoichiometric ecotoxicology and connect biological scales. AOPs have been

developed to conceptualize the knowledge linking a molecular initiating event of a

contaminant to an adverse outcome at a relevant level of biological organization via key

events (KE) and key event relationships. They can be used to link molecular responses to

underlying biological changes, an area of future work identified in figure 2. KEs can

1   occur at any level of biological organization and can be connected to DEB processes

2   (Murphy et al. 2018a; Murphy et al. 2018b). Traditional AOPs are chemically agnostic

3   and encode causal, not quantitative, relationships. However, the development of

4   quantitative AOPs (qAOPs) is an emerging field (Perkins et al., 2019). In qAOPs,

5   quantitative key event relationships, such as dose-response and response-response

6   relationships, can be influenced by ecotoxicological context through modulating factors

7   (Conolly et al. 2017). Modulating factors alter quantitative key event relationship

8   between two KEs, such as to change the shape of a response-response relationship.

9   Examples of modulating factors include the impact of environmental, genetic, disease, or

10   nutritional variations on the KE relationship (Villeneuve et al. 2014). Informing

11   appropriate modulating factors by nutrient-chemical interactions and relationships studied

12   in stoichiometric ecotoxicology will serve to refine quantitative relationships in qAOPs.

13   Connecting the established frameworks of DEB modeling and AOPs in circumstances

14   where elements might behave as stressors (under nutrient limitation or in excess) or

15   where contaminants behave as nutrients (e.g., hormesis at low doses) could improve the

16   classification, down-stream modeling, and prediction of emergent properties.

17   Future work is also needed to examine potential feedbacks among nutrient and

18   contaminant exposures at different levels of biological organization. For example,

19   contaminant exposure at the organismal and population level may influence element

20   cycling at the ecosystem level by changing organism physiological processes (e.g.,

21   growth and excretion). In turn, element cycling may also impact contaminant toxicity,

22   through somatic growth dilution (SGD) and reductions in bioaccumulation (Karimi et al.

23   2007). To date, very little is known about the extent, strength, and type of such feedbacks

and the circumstances under which they may occur. Such questions are particularly

important to address to understand changes in contaminant and element inputs to

ecosystems, including increases in nutrient pollution to aquatic systems.

There is also a need to examine the combined effects of element and contaminant

exposure on organisms across age, size, or life-stages. Mathematical models have been

used to incorporate stage structure into population dynamics. Structured population

models are able to incorporate additional complexity related to an additional variable,

such as age, size, or life-stage. These come in the form of discrete matrix equations

(Leslie, 1947), systems of ordinary differential equations, or continuous partial

differential equations (Sinko and Streifer 1967). Within the context of ecotoxicology,

systems of differential equations may include multiple compartments representing

various stages of the life cycle and, correspondingly, different effects of toxicants on the

stages of the life cycle. These models can include density-dependent effects of

populations. It is straightforward to incorporate ecotoxicological effects for the various

life-stages in a matrix model (Emlen and Springman 2007; Erickson et al. 2014) or a

system dynamics model (Weller et al. 2014). Furthermore, these models can easily be

extended to model nutritional stress (Weller et al. 2014; Hanson and Stark 2011; Caswell

2006).

Continuously-structured population models can be useful as they are more

computationally stable when estimating parameters that are dependent on time or the

structured variable. There are many continuously structured population models that

incorporate food availability (Ananthasubramaniam et al. 2011), and some that implicitly

consider food quality (Nisbet and Gurney 1983). However, explicit modeling of

1 stoichiometric constraints remains relatively unexplored within this framework.

2 Ecotoxicological effects have also largely remained un-integrated with continuously-

3 structured population models. Recently, however, Huang and Wang (2016) proposed a

4 toxin-mediated size-structured population model allowing for different sensitivities to

5 toxicants depending on size, age, etc.

6

7 **Incorporating stoichiometric ecotoxicology into management decisions.** Advancing

8 stoichiometric ecotoxicology promises to reduce uncertainty during environmental risk

9 assessment and management activities. This may be particularly true for extrapolations

10 across laboratory to field scales of biological organization, which remain among the

11 biggest research needs to achieve a more sustainable environment (Furley et al. 2018;

12 Van den Brink et al. 2018). Brooks, Fulton, and Hanson (2015) recommended accounting

13 for stoichiometric influences on aquatic ecotoxicology studies with plants and algae by

14 balancing experimental pragmatism with environmental realism. Traditional

15 ecotoxicology assays were developed to ensure sufficient growth of primary producers,

16 which represents a common endpoint to understand adverse outcomes of contaminant

17 exposures. To predict real-world outcomes, future research on ecotoxicology needs to be

18 more representative of field-relevant elemental gradients. If nutrient assimilation and

19 elemental ratios of lower trophic levels are modified, those changes will cascade into

20 changes in interspecific competition, the efficiency of secondary production, and

21 community composition.

22 　　We also propose better development and use of knowledge about how

23 stoichiometric conditions influence the production of natural toxins by aquatic and

24 terrestrial organisms. This is of particular relevance to ecological risk assessments in

1   inland and coastal waters, where harmful algal blooms (HAB) can elicit profound

2   impacts on aquatic life and public health and often surpass anthropogenic chemical risks.

3   In fact, HABs are now recognized as transformative threats to water quality and

4   confounders of environmental assessment and management efforts, which often rely on

5   risk-based regulatory frameworks for the protection of public health and the environment

6   (Brooks et al. 2016). Whereas ecological studies and monitoring activities have

7   previously examined toxin concentrations, these efforts are routinely limited by the

8   absence of robust analytical quantitation of diverse toxins produced by specific HAB

9   species. This represents a critical consideration for water resource management because

10   HAB forming species are assumed to be producing toxins even if measured toxins are

11   below detection limits. These conservative measures decrease the exposure of toxins but

12   may come at an unnecessary cost. While water quality models can predict the occurrence,

13   intensity, and severity of HABs, by not incorporating toxin production these models

14   cannot predict the risks to human or ecosystem health. Developing predictive growth,

15   toxin production, and comparative toxicity models across stoichiometric nutrient

16   gradients are imperative for forecasting, diagnosing, and preventing ecological and

17   human health risks presented by algal toxins.

18
19   **Concluding Remarks**

20   Here we refer to the integration of ecotoxicology and ES theory as stoichiometric

21   ecotoxicology. The conceptual framework for stoichiometric ecotoxicology that we have

22   developed highlights the interactive impacts of elemental imbalances and chemical

23   contaminants within and across scales from sub-organismal to ecosystems. This

24   conceptual framework was built from existing empirical and theoretical examples that

1   link stoichiometry and toxicology. While an increasing amount of stoichiometrically

2   explicit models have provided insight into ecological interactions, there is much to be

3   gained by incorporating concurrent nutrient and toxicant stressors into models formulated

4   under the presented stoichiometric ecotoxicology framework. This unifying framework

5   offers the potential to deepen mechanistic understandings of the adverse outcomes of

6   chemicals across ecological scales and improve the predictive powers of

7   ecotoxicology.

8

9

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15

16

References Cited

18 Altenburger, R., W. Boedeker, M. Faust, and L. H. Grimme. 1996. "Regulations for
19   combined effects of pollutants: Consequences from risk assessment in aquatic
20   toxicology." *Food and Chemical Toxicology* 34:1155-1157.
21 Ananthasubramaniam, Bharath, Roger M. Nisbet, William A. Nelson, Edward McCauley,
22   and William S. C. Gurney. 2011. "Stochastic growth reduces population fluctuations
23   in *Daphnia*-algal systems." *Ecology* 92:362-372.
24 Andersen, Tom. 1997. *Pelagic nutrient cycles; herbivores as sources and sinks.*
25   Springer,Berlin.
26 Andersen, Tom, James J. Elser, and Dag O. Hessen. 2004. "Stoichiometry and population
27   dynamics." *Ecology Letters* 7:884-900.
28 Anderson, Thomas R. and Dag O. Hessen. 2005. "Threshold elemental ratios for carbon
29   versus phosphorus limitation in *Daphnia*." *Freshwater Biology* 50:2063-2075.
30 Anderson, Thomas R., David Raubenheimer, Dag O. Hessen, Kim Jensen, Windy C.
31   Gentleman, and Daniel J. Mayor. 2020. "Unifying Concepts of Animal Nutrition to

Understand How Protein-Rich Diets Can Be "Too Much of a Good Thing." *Front. Ecol. Evol.* 8(196).

Ankley, Gerald T., Richard S. Bennett, Russell J. Erickson, Dale J. Hoff, Michael W. Hornung, Rodney D. Johnson, David R. Mount, John W. Nichols, Christine L. Russom, Patricia K. Schmieder, and others. 2010. "Adverse outcome pathways: a conceptual framework to support ecotoxicology research and risk assessment." *Environmental Toxicology and Chemistry: An International Journal* 29:730-741.

Ankley, Gerald T., Russell J. Erickson, Gary L. Phipps, Vincent R. Mattson, Patricia A. Kosian, Barbara R. Sheedy, and Julie S. Cox. 1995. "Effects of light intensity on the phototoxicity of fluoranthene to a benthic macroinvertebrate." *Environmental science & technology* 29:2828-2833.

Arce-Funck, J., H. Clivot, V. Felten, P. Rousselle, F. Guérold, and M. Danger. 2013. "Phosphorus availability modulates the toxic effect of silver on aquatic fungi and leaf litter decomposition." *Aquatic toxicology* 144:199-207.

Arce-Funck, Julio, Clément Crenier, Michael Danger, Elise Billoir, Philippe Usseglio-Polatera, and Vincent Felten. 2018. "High stoichiometric food quality increases moulting organism vulnerability to pollutant impacts: an experimental test with *Gammarus fossarum* (*Crustacea: Amphipoda*)." *Science of The Total Environment* 645:1484-1495.

Arce-Funck, Julio, Clément Crenier, Michael Danger, Carole Cossu-Leguille, François Guérold, and Vincent Felten. 2016. "Stoichiometric constraints modulate impacts of silver contamination on stream detritivores: an experimental test with *Gammarus fossarum*." *Freshwater biology* 61:2075-2089.

Arnot, Jon A. and Frank A. Gobas. 2004. "A food web bioaccumulation model for organic chemicals in aquatic ecosystems." *Environmental Toxicology and Chemistry: An International Journal* 23(10):2343-2355.

Arnot, Jon A. and Frank A. Gobas. 2006. "A review of bioconcentration factor (BCF) and bioaccumulation factor (BAF) assessments for organic chemicals in aquatic organisms.." *Environmental Reviews* 14(4):257-297.

Ashauer, Roman, Alistair B. A. Boxall, and Colin D. Brown. 2007. "New ecotoxicological model to simulate survival of aquatic invertebrates after exposure to fluctuating and sequential pulses of pesticides." *Environmental science & technology* 41:1480-1486.

Ashauer, Roman and Colin D. Brown. 2008. "Toxicodynamic assumptions in ecotoxicological hazard models." *Environmental Toxicology and Chemistry* 27:1817-1821.

Back, Jeffrey A. and Ryan S. King. 2013. "Sex and size matter: ontogenetic patterns of nutrient content of aquatic insects." *Freshwater Science* 32:837-848.

Baird, Donald J., Lorraine Maltby, Peter W. Greig-Smith, and Peter E. T. Douben 1996. "Putting the 'ECO-'into ECOtoxicology." Pp. 1-4 in *ECOtoxicology: Ecological Dimensions*. Springer.

Banerjee, Malay and Ezio Venturino. 2011. "A phytoplankton-toxic phytoplankton-zooplankton model." *Ecological Complexity* 8:239-248.

Baudrot, Virgile, Clémentine Fritsch, Antoine Perasso, Malay Banerjee, and Francis Raoul. 2018. "Effects of contaminants and trophic cascade regulation on food chain stability: Application to cadmium soil pollution on small mammals-Raptor systems." *Ecological Modelling* 382:33-42.

1 Baxter, Leilan, Richard A. Brain, Linda Lissemore, Keith R. Solomon, Mark L. Hanson,
2   and Ryan S. Prosser. 2016. "Influence of light, nutrients, and temperature on the
3   toxicity of atrazine to the algal species *Raphidocelis subcapitata*: implications for the
4   risk assessment of herbicides." *Ecotoxicology and environmental safety* 132:250-259.
5 Bian, Jingyi, Jason P. Berninger, Barry A. Fulton, and Bryan W. Brooks. 2013. "Nutrient
6   stoichiometry and concentrations influence silver toxicity in the aquatic macrophyte
7   *Lemna gibba*." *Science of the Total Environment* 449:229-236.
8 Bontje, D., B. W. Kooi, M. Liebig, and S. A. L. M. Kooijman. 2009. "Modelling long-term
9   ecotoxicological effects on an algal population under dynamic nutrient stress." *water
10   research* 43:3292-3300.
11 Borgå, Katrine, Karen A. Kidd, Derek C. Muir, Olof Berglund, Jason M. Conder, Frank A.
12   Gobas, John Kucklick, Olaf Malm, and David E. Powell. 2014. "Trophic magnification
13   factors: considerations of ecology, ecosystems, and study design." *Integrated
14   environmental assessment and management* 8(1):64-84.
15 Brooks, Bryan W., Barry A. Fulton, and Mark L. Hanson. 2015. "Aquatic toxicology
16   studies with macrophytes and algae should balance experimental pragmatism with
17   environmental realism." *Science of the Total Environment* 536:406-407.
18 Brooks, Bryan W., James M. Lazorchak, Meredith D. A. Howard, Mari-Vaughn V.
19   Johnson, Steve L. Morton, Dawn A. K. Perkins, Euan D. Reavie, Geoffrey I. Scott,
20   Stephanie A. Smith, and Jeffery A. Steevens. 2016. "Are harmful algal blooms
21   becoming the greatest inland water quality threat to public health and aquatic
22   ecosystems?" *Environmental toxicology and chemistry* 35:6-13.
23 Bryant, John P., F. S. Chapin III, and David R. Klein. 1983. "Carbon/nutrient balance of
24   boreal plants in relation to vertebrate herbivory." *Oikos* 357-368.
25 Bullejos, Francisco J., Presentación Carrillo, Elena Gorokhova, Juan M. Medina-Sánchez,
26   Esteban G. Balseiro, and Manuel Villar-Argaiz. 2014. "Shifts in food quality for
27   herbivorous consumer growth: multiple golden means in the life history." *Ecology*
28   95:1272-1284.
29 Bundschuh, Mirco, Torsten Hahn, Mark O. Gessner, and Ralf Schulz. 2009. "Antibiotics
30   as a chemical stressor affecting an aquatic decomposer-detritivore system."
31   *Environmental Toxicology and Chemistry: An International Journal* 28:197-203.
32 Cairns, John. 1983. "Are single species toxicity tests alone adequate for estimating
33   environmental hazard?" *Hydrobiologia* 100:47-57.
34 Cairns, John. 1984. "Are single species toxicity tests alone adequate for estimating
35   environmental hazard?" *Environmental Monitoring and Assessment* 4:259-273.
36 Cairns Jr, John. 1988. "Putting the eco in ecotoxicology." *Regulatory Toxicology and
37   Pharmacology* 8:226-238.
38 Calabrese, Edward J. and Linda A. Baldwin. 2002. "Defining hormesis." *Human &
39   experimental toxicology* 21:91-97.
40 Calabrese, Edward J. and Mark P. Mattson. 2017. "How does hormesis impact biology,
41   toxicology, and medicine?" *NPJ aging and mechanisms of disease* 3:13.
42 Calow, Peter 1996. "Ecology in ecotoxicology: some possible 'rules of thumb'." Pp. 5-12
43   in *ECOtoxicology: ecological dimensions*. Springer.
44 Caswell, Hal. 2006. "Matrix population models." *Encyclopedia of Environmetrics* 3.

Cherchi, Carla, Milos Miljkovic, Max Diem, and April Z. Gu. 2015. "nTiO$_2$ induced changes in intracellular composition and nutrient stoichiometry in primary producer—cyanobacteria." *Science of the Total Environment* 512:345-352.

Chiou, Cary T. 1985. "Partition coefficients of organic compounds in lipid-water systems and correlations with fish bioconcentration factors." *Environmental science & technology* 19:57-62.

Clivot, Hugues, Faustine Charmasson, Vincent Felten, Jean-Pierre Boudot, François Guérold, and Michael Danger. 2014. "Interactive effects of aluminium and phosphorus on microbial leaf litter processing in acidified streams: A microcosm approach." *Environmental pollution* 186:67-74.

Conine, Andrea L. and Paul C. Frost. 2017. "Variable toxicity of silver nanoparticles to *Daphnia magna*: effects of algal particles and animal nutrition." *Ecotoxicology* 26:118-126.

Conolly, Rory B., Gerald T. Ankley, WanYun Cheng, Michael L. Mayo, David H. Miller, Edward J. Perkins, Daniel L. Villeneuve, and Karen H. Watanabe. 2017. "Quantitative adverse outcome pathways and their application to predictive toxicology." *Environmental science & technology* 51(8):4661-4672.

Costello, David M., Konrad J. Kulacki, Mary E. McCarthy, Scott D. Tiegs, and Bradley J. Cardinale. 2018. "Ranking stressor impacts on periphyton structure and function with mesocosm experiments and environmental-change forecasts." *PloS one* 13:e0204510.

Cross, Wyatt F., Jonathan P. Benstead, Amy D. Rosemond, and J. Bruce Wallace. 2003. "Consumer-resource stoichiometry in detritus-based streams." *Ecology Letters* 6:721-732.

Cunning, Ross, Erik B. Muller, Ruth D. Gates, and Roger M. Nisbet. 2017. "A dynamic bioenergetic model for coral-Symbiodinium symbioses and coral bleaching as an alternate stable state." *Journal of theoretical biology* 431:49-62.

Danger, Michael and Florence Maunoury-Danger 2013. "Ecological Stoichiometry." Pp. 317-326 in *Encyclopedia of Aquatic Ecotoxicology*. Springer.

Darchambeau, François, Per J. Faerøvig, and Dag O. Hessen. 2003. "How *Daphnia* copes with excess carbon in its food." *Oecologia* 136:336-346.

Das, Pranab, Chris D. Metcalfe, and Marguerite A. Xenopoulos. 2014. "Interactive effects of silver nanoparticles and phosphorus on phytoplankton growth in natural waters." *Environmental science & technology* 48:4573-4580.

Droop, M. R. 1974. "The nutrient status of algal cells in continuous culture." *Journal of the Marine Biological Association of the United Kingdom* 54:825-855.

Elser, James J., Thomas H. Chrzanowski, Robert W. Sterner, and Kenneth H. Mills. 1998. "Stoichiometric constraints on food-web dynamics: a whole-lake experiment on the Canadian Shield." *Ecosystems* 1:120-136.

Elser, James J., Marcia Kyle, Jennifer Learned, Michelle L. McCrackin, Angela Peace, and Laura Steger. 2016. "Life on the stoichiometric knife-edge: effects of high and low food C: P ratio on growth, feeding, and respiration in three *Daphnia* species." *Inland Waters* 6:136-146.

Elser, James J., Irakli Loladze, Angela L. Peace, and Yang Kuang. 2012. "Lotka re-loaded: modeling trophic interactions under stoichiometric constraints." *Ecological Modelling* 245:3-11.

Elser, J. J., R. W. Sterner, E. Gorokhova, W. F. Fagan, T. A. Markow, J. B. Cotner, J. F. Harrison, S. E. Hobbie, G. M. Odell, and L. W. Weider. 2000. "Biological stoichiometry from genes to ecosystems." *Ecology Letters* 3:540-550.

Elser, James J. and Jotaro Urabe. 1999. "The stoichiometry of consumer-driven nutrient recycling: theory, observations, and consequences." *Ecology* 80:735-751.

Emlen, John M. and Kathrine R. Springman. 2007. "Developing methods to assess and predict the population level effects of environmental contaminants." *Integrated Environmental Assessment and Management: An International Journal* 3:157-165.

Erickson, Richard A., Stephen B. Cox, Jessica L. Oates, Todd A. Anderson, Christopher J. Salice, and Kevin R. Long. 2014. "A *Daphnia* population model that considers pesticide exposure and demographic stochasticity." *Ecological modelling* 275:37-47.

Evans-White, Michelle A., Walter K. Dodds, Donald G. Huggins, and Debra S. Baker. 2009. "Thresholds in macroinvertebrate biodiversity and stoichiometry across water-quality gradients in Central Plains (USA) streams." *Journal of the North American Benthological Society* 28:855-868.

Fernandes, Isabel, Sofia Duarte, Fernanda Cássio, and Cláudia Pascoal. 2009. "Mixtures of zinc and phosphate affect leaf litter decomposition by aquatic fungi in streams." *Science of the total environment* 407:4283-4288.

Forbes, Valery E., Chris J. Salice, Bjorn Birnir, Randy J. F. Bruins, Peter Calow, Virginie Ducrot, Nika Galic, Kristina Garber, Bret C. Harvey, Henriette Jager, and others. 2017. "A framework for predicting impacts on ecosystem services from (sub) organismal responses to chemicals." *Environmental toxicology and chemistry* 36:845-859.

Frost, Paul C., Jonathan P. Benstead, Wyatt F. Cross, Helmut Hillebrand, James H. Larson, Marguerite A. Xenopoulos, and Takehito Yoshida. 2006. "Threshold elemental ratios of carbon and phosphorus in aquatic consumers." *Ecology Letters* 9:774-779.

Frost, P. C. and J. J. Elser. 2002. "Growth responses of littoral mayflies to the phosphorus content of their food." *Ecology Letters* 5:232-240.

Frost, Paul C., Michelle A. Evans-White, Zoe V. Finkel, Thomas C. Jensen, and Virginia Matzek. 2005. "Are you what you eat? Physiological constraints on organismal stoichiometry in an elementally imbalanced world." *Oikos* 109:18-28.

Frost, Paul C. and Nancy C. Tuchman. 2005. "Nutrient release rates and ratios by two stream detritivores fed leaf litter grown under elevated atmospheric CO_2." *Archiv für Hydrobiologie* 163:463-477.

Frost, Paul C., Marguerite A. Xenopoulos, and James H. Larson. 2004. "The stoichiometry of dissolved organic carbon, nitrogen, and phosphorus release by a planktonic grazer, *Daphnia*." *Limnology and Oceanography* 49:1802-1808.

Fulton, Barry A., Richard A. Brain, Sascha Usenko, Jeffrey A. Back, and Bryan W. Brooks. 2010. "Exploring *Lemna gibba* thresholds to nutrient and chemical stressors: differential effects of triclosan on internal stoichiometry and nitrate uptake across a nitrogen: phosphorus gradient." *Environmental Toxicology and Chemistry* 29:2363-2370.

Fulton, Barry A., Richard A. Brain, Sascha Usenko, Jeffrey A. Back, Ryan S. King, and Bryan W. Brooks. 2009. "Influence of nitrogen and phosphorus concentrations and ratios on *Lemna gibba* growth responses to triclosan in laboratory and stream mesocosm experiments." *Environmental toxicology and chemistry* 28:2610-2621.

Furley, Tatiana H., Julie Brodeur, Helena C. Silva de Assis, Pedro Carriquiriborde, Katia R. Chagas, Jone Corrales, Marina Denadai, Julio Fuchs, Renata Mascarenhas, Karina S. B. Miglioranza, and others. 2018. "Toward sustainable environmental quality: Identifying priority research questions for Latin America." *Integrated environmental assessment and management* 14:344-357.

Galic, Nika, Volker Grimm, and Valery E. Forbes. 2017. "Impaired ecosystem process despite little effects on populations: modeling combined effects of warming and toxicants." *Global change biology* 23:2973-2989.

Galic, Nika, Lauren L. Sullivan, Volker Grimm, and Valery E. Forbes. 2018. "When things don't add up: quantifying impacts of multiple stressors from individual metabolism to ecosystem processing." *Ecology letters* 21:568-577.

Garner, Kendra L., Sangwon Suh, Hunter S. Lenihan, and Arturo A. Keller. 2015. "Species sensitivity distributions for engineered nanomaterials." *Environmental science & technology* 49:5753-5759.

Gessner, Mark O. and Ahmed Tlili. 2016. "Fostering integration of freshwater ecology with ecotoxicology." *Freshwater biology* 61:1991-2001.

Geyer, Harald J., Irene Scheunert, Rainer Bruggemann, Michael Matthies, Christian E. W. Steinberg, Vladimir Zitko, Antonius Kettrup, and Wayne Garrison. 1994. "The relevance of aquatic organisms lipid content to the toxicity of lipophilic chemicals: Toxicity of lindane to different fish species." *Ecotoxicology and environmental safety* 28:53-70.

Gobas, Frank A. 1993. "A model for predicting the bioaccumulation of hydrophobic organic chemicals in aquatic food-webs: application to Lake Ontario." *Ecological Modelling* 69(1-2):1-17.

Gonçalves, Renata A., Ana L. d. O. F. Rossetto, Diego J. Nogueira, Denice S. Vicentini, and William G. Matias. 2018. "Comparative assessment of toxicity of ZnO and amine-functionalized ZnO nanorods toward *Daphnia magna* in acute and chronic multigenerational tests." *Aquatic toxicology* 197:32-40.

Grover, James P. 2002. "Stoichiometry, herbivory and competition for nutrients: simple models based on planktonic ecosystems." *Journal of theoretical biology* 214:599-618.

Grover, James P. 2003. "The impact of variable stoichiometry on predator-prey interactions: a multinutrient approach." *The American Naturalist* 162:29-43.

Grover, James P. 2004. "Predation, competition, and nutrient recycling: a stoichiometric approach with multiple nutrients." *Journal of theoretical biology* 229:31-43.

Grzesiuk, Malgorzata, Alexander Wacker, and Elly Spijkerman. 2016. "Photosynthetic sensitivity of phytoplankton to commonly used pharmaceuticals and its dependence on cellular phosphorus status." *Ecotoxicology* 25:697-707.

Hallam, T. G. and J. T. De Luna. 1984. "Effects of toxicants on populations: A qualitative: Approach III. Environmental and food chain pathways." *Journal of Theoretical Biology* 109:411-429.

Hansen, Laura K., Paul C. Frost, James H. Larson, and Chris D. Metcalfe. 2008. "Poor elemental food quality reduces the toxicity of fluoxetine on *Daphnia magna*." *Aquatic toxicology* 86:99-103.

Hanson, Niklas and John D. Stark. 2011. "An approach for developing simple generic models for ecological risk assessments of fish populations." *Environmental toxicology and chemistry* 30:2372-2383.

Hassan, Md N., Kelsey Thompson, Gregory Mayer, and Angela Peace. 2018. "Effect of excess food nutrient on producer-grazer model under stoichiometric and toxicological constraints." *Mathematical biosciences and engineering: MBE* 16:150-167.

Hessen, Dag O., James J. Elser, Robert W. Sterner, and Jotato Urabe. 2013. "Ecological stoichiometry: an elementary approach using basic principles." *Limnology and Oceanography* 58(6):2219-2236.

Huang, Qihua and Hao Wang. 2016. "A toxin-mediated size-structured population model: Finite difference approximation and well-posedness." *Mathematical Biosciences & Engineering* 13:697-722.

Huang, Qihua, Hao Wang, and Mark A. Lewis. 2014. "Development of a Toxin-Mediated Predator-Prey Model Applicable to Aquatic Environments in the Athabasca Oil Sands Region.".

Huang, Qihua, Hao Wang, and Mark A. Lewis. 2015. "The impact of environmental toxins on predator-prey dynamics." *Journal of theoretical biology* 378:12-30.

Ieromina, Oleksandra, Willie J. G. M. Peijnenburg, Geert Snoo, Jutta Müller, Thomas P. Knepper, and Martina G. Vijver. 2014. "Impact of imidacloprid on *Daphnia magna* under different food quality regimes." *Environmental toxicology and chemistry* 33:621-631.

Jager, Tjalling. 2013. "Making sense of chemical stress." *VU University*.

Jager, T. 2016. "Dynamic Modeling for Uptake and Effects of Chemicals." Pp. 71-98 in *Marine Ecotoxicology*. Elsevier.

Jager, Tjalling, Alpar Barsi, Natnael T. Hamda, Benjamin T. Martin, Elke I. Zimmer, and Virginie Ducrot. 2014. "Dynamic energy budgets in population ecotoxicology: Applications and outlook." *Ecological Modelling* 280:140-147.

Jager, Tjalling, Eva M. Gudmundsdottir, and Nina Cedergreen. 2014. "Dynamic modeling of sublethal mixture toxicity in the nematode *Caenorhabditis elegans*." *Environmental science & technology* 48:7026-7033.

Jager, Tjalling, Evelyn H. W. Heugens, and Sebastiaan A. L. M. Kooijman. 2006. "Making sense of ecotoxicological test results: towards application of process-based models." *Ecotoxicology* 15:305-314.

Jager, Tjalling and Elke I. Zimmer. 2012. "Simplified dynamic energy budget model for analysing ecotoxicity data." *Ecological Modelling* 225:74-81.

Janssens, Lizanne, Lin Op de Beeck, and Robby Stoks. 2016. "Stoichiometric responses to an agricultural pesticide are modified by predator cues." *Environmental science & technology* 51:581-588.

Jiang, Jie, Anglu Shen, Hao Wang, and Sanling Yuan. 2019. "Regulation of phosphate uptake kinetics in the bloom-forming dinoflagellates *prorocentrum donghaiense* with emphasis on two-stage dynamic process." *Journal of theoretical biology* 463:12-21.

Karimi, R., F. N. S. a. F. C. L. 2010. "Multielement stoichiometry in aquatic invertebrates: when growth dilution matters." *The American Naturalist* 127(6):699-709.

1 Karimi, Roxanne, Celia Y. Chen, Paul C. Pickhardt, Nicholas S. Fisher, and Carol L. Folt.
2   2007. "Stoichiometric controls of mercury dilution by growth." *Proceedings of the*
3   *National Academy of Sciences* 104:7477-7482.
4 Karimi, Roxanne and Carol L. Folt. 2006. "Beyond macronutrients: element variability
5   and multielement stoichiometry in freshwater invertebrates." *Ecology letters*
6   9:1273-1283.
7 Kearney, Michael, Stephen J. Simpson, David Raubenheimer, and Brian Helmuth. 2010.
8   "Modelling the ecological niche from functional traits." *Philosophical Transactions of*
9   *the Royal Society B: Biological Sciences* 365:3469-3483.
10 Kilham, Susan S. 1998. "Effects of physiological state on the bioaccumulation of toxic
11   chemicals in algae and their transfer to zooplankton." *Internationale Vereinigung für*
12   *theoretische und angewandte Limnologie: Verhandlungen* 26:1734-1736.
13 Klanjscek, Tin, Roger M. Nisbet, John H. Priester, and Patricia A. Holden. 2012.
14   "Modeling physiological processes that relate toxicant exposure and bacterial
15   population dynamics." *PLoS One* 7:e26955.
16 Klanjscek, Tin, Roger M. Nisbet, John H. Priester, and Patricia A. Holden. 2013.
17   "Dynamic energy budget approach to modeling mechanisms of CdSe quantum dot
18   toxicity." *Ecotoxicology* 22:319-330.
19 Kooijman, S. A. L. M. 1998. "The synthesizing unit as model for the stoichiometric
20   fusion and branching of metabolic fluxes." *Biophysical chemistry* 73:179-188.
21 Kooijman, Sebastiaan A. L. M. 1998. "The synthesizing unit as model for the
22   stoichiometric fusion and branching of metabolic fluxes." *Biophysical chemistry*
23   73:179-188.
24 Kooijman, S. A. L. M., T. Andersen, and B. W. Kooi. 2004. "Dynamic energy budget
25   representations of stoichiometric constraints on population dynamics." *Ecology*
26   85:1230-1243.
27 Kooijman, S. A. L. M. and J. J. M. Bedaux. 1996. "Analysis of toxicity tests on *Daphnia*
28   survival and reproduction." *Water Research* 30:1711-1723.
29 Kooijman, Bas and S. A. L. M. Kooijman. 2010. *Dynamic energy budget theory for*
30   *metabolic organisation*. Cambridge university press.
31 Kopácek, Jirı, Josef Hejzlar, Jakub Borovec, Petr Porcal, and Ivana Kotorová. 2000.
32   "Phosphorus inactivation by aluminum in the water column and sediments:
33   Lowering of in-lake phosphorus availability in an acidified watershed-lake
34   ecosystem." *Limnology and Oceanography* 45:212-225.
35 Kuijper, Lothar D. J., Thomas R. Anderson, and Sebastiaan A. L. M. Kooijman. 2004b. "C
36   and N gross growth efficiencies of copepod egg production studied using a Dynamic
37   Energy Budget model." *Journal of Plankton Research* 26:213-226.
38 Kuijper, Lothar D. J., Bob W. Kooi, Thomas R. Anderson, and Sebastiaan A. L. M.
39   Kooijman. 2004a. "Stoichiometry and food-chain dynamics." *Theoretical Population*
40   *Biology* 66:323-339.
41 Kwak, Kyunghee, Kyunghee Ji, Younglim Kho, Pilje Kim, Jaean Lee, Jisung Ryu, and
42   Kyungho Choi. 2018. "Chronic toxicity and endocrine disruption of naproxen in
43   freshwater waterfleas and fish, and steroidogenic alteration using H295R cell
44   assay." *Chemosphere* 204:156-162.
45 Lessard, Charlotte R. and Paul C. Frost. 2012. "Phosphorus nutrition alters herbicide
46   toxicity on *Daphnia magna*." *Science of the total environment* 421:124-128.

Liu, Ying, Jian Zhang, Baoyu Gao, and Suping Feng. 2014. "Combined effects of two antibiotic contaminants on *Microcystis aeruginosa*." *Journal of Hazardous Materials* 279:148-155.

Logan, J. D., Anthony Joern, and William Wolesensky. 2004. "Mathematical model of consumer homeostasis control in plant-herbivore dynamics." *Mathematical and computer modelling* 40:447-456.

Loladze, Irakli, Yang Kuang, and James J. Elser. 2000. "Stoichiometry in producer-grazer systems: linking energy flow with element cycling." *Bulletin of Mathematical Biology* 62:1137-1162.

Loladze, I., Y. Kuang, J. J. Elser, and W. F. Fagan. 2004. "Coexistence of two predators on one prey mediated by stoichiometry." *Theoretical Population Biology* 65:1-15.

Mackay, D., H. Puig, and L. S. McCarty. 1992. "An equation describing the time course and variability in uptake and toxicity of narcotic chemicals to fish." *Environmental toxicology and chemistry* 11:941-951.

Marques, Gonçalo M., Starrlight Augustine, Konstadia Lika, Laure Pecquerie, Tiago Domingos, and Sebastiaan A. L. M. Kooijman. 2018. "The AmP project: comparing species on the basis of dynamic energy budget parameters." *PLoS computational biology* 14:e1006100.

Martin, B., T. Jager, R.M. Nisbet, T.G. Preuss, and V. Grimm. 2014. "Limitations of extrapolating toxic effects on reproduction to the population level." *Ecological applications* 24(8):1972-1983.

Martin, Benjamin T., Tjalling Jager, Roger M. Nisbet, Thomas G. Preuss, Monika Hammers-Wirtz, and Volker Grimm. 2013. "Extrapolating ecotoxicological effects from individuals to populations: a generic approach based on Dynamic Energy Budget theory and individual-based modeling." *Ecotoxicology* 22:574-583.

Mkandawire, Martin and E. G. Dudel. 2012. "Homeostatic regulation of elemental stoichiometry by *Lemna gibba* L. G3 when nutrient interact with toxic metals." *Ecotoxicology* 21:456-464.

Muller, Erik B., Sebastiaan A. L. M. Kooijman, Peter J. Edmunds, Francis J. Doyle, and Roger M. Nisbet. 2009. "Dynamic energy budgets in syntrophic symbiotic relationships between heterotrophic hosts and photoautotrophic symbionts." *Journal of Theoretical Biology* 259:44-57.

Muller, Erik B., Roger M. Nisbet, Sebastiaan A. L. M. Kooijman, James J. Elser, and Edward. McCauley. 2001. "Stoichiometric food quality and herbivore dynamics." *Ecology Letters* 4:519-529.

Murdock, J N., F D. Shields Jr., and R E. Lizotter Jr. 2013. "Periphyton responses to nutrients and atrazine introduced through agricultural runoff in riverine wetland." *Ecotoxicology* 22(2):215-230.

Murdock, J N. and D L. Wetzel. 2012. "Measuring the response of individual algal cells to nutrient and herbicide mixtures within natural biofilms." *Microbial Ecology* 63:761-722.

Murphy, Cheryl A., Roger M. Nisbet, Philipp Antczak, Natàlia Garcia-Reyero, Andre Gergs, Konstadia Lika, Teresa Mathews, Erik B. Muller, Diane Nacci, Angela Peace, and others 2018a. "Linking adverse outcome pathways to dynamic energy budgets: A conceptual model." Pp. 281-302 in *A Systems Biology Approach to Advancing Adverse Outcome Pathways for Risk Assessment*. Springer.

Murphy, Cheryl A., Roger M. Nisbet, Philipp Antczak, Natàlia Garcia-Reyero, Andre Gergs, Konstadia Lika, Teresa Mathews, Erik B. Muller, Diane Nacci, Angela Peace, and others. 2018b. "Incorporating suborganismal processes into dynamic energy budget models for ecological risk assessment." *Integrated environmental assessment and management* 14:615-624.

Newman, Michael C. and William H. Clements. 2007. *Ecotoxicology: a comprehensive treatment.* cRc Press.

Ni, Felicity J., Noreen E. Kelly, and George B. Arhonditsis. 2017. "Towards the development of an ecophysiological *Daphnia* model to examine effects of toxicity and nutrition." *Ecological informatics* 41:91-107.

Nisbet, R. M. and W. S. C. Gurney. 1983. "The systematic formulation of population models for insects with dynamically varying instar duration." *Theoretical Population Biology* 23:114-135.

Nisbet, R. M., E. B. Muller, K. Lika, and S. A. L. M. Kooijman. 2000. "From molecules to ecosystems through dynamic energy budget models." *Journal of animal ecology* 69:913-926.

Nuttens, A., S. Chatellier, S. Devin, C. Guignard, A. Lenouvel, and E. M. Gross. 2016. "Does nitrate co-pollution affect biological responses of an aquatic plant to two common herbicides?" *Aquatic Toxicology* 177:355-364.

Ockleford, C., P. Adriaanse, P. Berny, T. Brock, S. Duquesne, S. Grilli, A.F. Hernandez-Jerez, S.H. Bennekou, M. Klein, and T. Kuhl. 2018. "Scientific Opinion on the state of the art of Toxicokinetic/Toxicodynamic (TKTD) effect models for regulatory risk assessment of pesticides for aquatic organisms." *EFSA Journal* 16(8).

Peace, Angela. 2015. "Effects of light, nutrients, and food chain length on trophic efficiencies in simple stoichiometric aquatic food chain models." *Ecological Modelling* 312:125-135.

Peace, Angela, Monica D. Poteat, and Hao Wang. 2016. "Somatic growth dilution of a toxicant in a predator-prey model under stoichiometric constraints." *Journal of theoretical biology* 407:198-211.

Peace, Angela, Hao Wang, and Yang Kuang. 2014. "Dynamics of a Producer-Grazer Model Incorporating the Effects of Excess Food Nutrient Content on Grazer's Growth." *Bulletin of mathematical biology* 76:2175-2197.

Peace, Angela, Yuqin Zhao, Irakli Loladze, James J. Elser, and Yang Kuang. 2013. "A stoichiometric producer-grazer model incorporating the effects of excess food-nutrient content on consumer dynamics." *Mathematical biosciences*.

Pecquerie, Laure, Roger M. Nisbet, Ronan Fablet, Anne Lorrain, and Sebastiaan A. L. M. Kooijman. 2010. "The impact of metabolism on stable isotope dynamics: a theoretical framework." *Philosophical Transactions of the Royal Society B: Biological Sciences* 365:3455-3468.

Phillips, Donald L. and Peter M. Eldridge. 2006. "Estimating the timing of diet shifts using stable isotopes." *Oecologia* 147(2):195-203.

Pieters, Barry J., Tjalling Jager, Michiel H. S. Kraak, and Wim Admiraal. 2006. "Modeling responses of *Daphnia magna* to pesticide pulse exposure under varying food conditions: intrinsic versus apparent sensitivity." *Ecotoxicology* 15:601-608.

Rana, Md M., Chandani Dissanayake, Lourdes Juan, Kevin R. Long, and Angela Peace. 2018. "Mechanistically derived spatially heterogeneous producer-grazer model

subject to stoichiometric constraints." *Mathematical biosciences and engineering: MBE* 16:222-233.

Raubenheimer, David. 2011. "Toward a quantitative nutritional ecology: the right-angled mixture triangle." *Ecological Monographs* 81(3):407-427.

Raubenheimer, David, Steven J. Simpson, and David Mayntz. 2009. "Nutrition, ecology and nutritional ecology: toward an integrated framework." *Functional Ecology* 4-16.

Relyea, Rick and Jason Hoverman. 2006. "Assessing the ecology in ecotoxicology: a review and synthesis in freshwater systems." *Ecology Letters* 9:1157-1171.

Rivas-Ubach, Albert, Jordi Sardans, Miriam Pérez-Trujillo, Marc Estiarte, and Josep Peñuelas. 2012. "Strong relationship between elemental stoichiometry and metabolome in plants." *Proceedings of the National Academy of Sciences* 109:4181-4186.

Rosi-Marshall, Emma J. and Todd V. Royer. 2012. "Pharmaceutical compounds and ecosystem function: an emerging research challenge for aquatic ecologists." *Ecosystems* 15:867-880.

Santore, Robert C., Adam C. Ryan, Frode Kroglund, Patricio H. Rodriguez, William A. Stubblefield, Allison S. Cardwell, William J. Adams, and Eirik Nordheim. 2018. "Development and application of a biotic ligand model for predicting the chronic toxicity of dissolved and precipitated aluminum to aquatic organisms." *Environmental toxicology and chemistry* 37:70-79.

Sinko, James W. and William Streifer. 1967. "A new model for age-size structure of a population." *Ecology* 48:910-918.

Sousa, Tânia, Tiago Domingos, J.-C. Poggiale, and S. A. L. M. Kooijman 2010. "Dynamic energy budget theory restores coherence in biology." The Royal Society.

Souza, Jaqueline P., Francine P. Venturini, Fabrício Santos, and Valtencir Zucolotto. 2018. "Chronic toxicity in Ceriodaphnia dubia induced by graphene oxide." *Chemosphere* 190:218-224.

Sperfeld, Erik, Nicole D. Wagner, Halvor M. Halvorson, Matthew Malishev, and David Raubenheimer. 2017. "Bridging ecological stoichiometry and nutritional geometry with homeostasis concepts and integrative models of organism nutrition." *Functional Ecology* 31:286-296.

Steinman, Alan D. and Mary Ogdahl. 2008. "Ecological effects after an alum treatment in Spring Lake, Michigan." *Journal of Environmental Quality* 37(1):22-19.

Sterner, Robert W. 1997. "Modelling interactions of food quality and quantity in homeostatic consumers." *Freshwater Biology* 38:473-481.

Sterner, Robert W. and James J. Elser. 2002. *Ecological stoichiometry: the biology of elements from molecules to the biosphere*. Princeton university press.

Stumm, W and JJ. Morgan. 2012. *Aquatic chemistry: chemical equilibria and rates in natural waters.* John Wiley & Sons.

Suzuki-Ohno, Yukari, Masakado Kawata, and Jotaro Urabe. 2012. "Optimal feeding under stoichiometric constraints: a model of compensatory feeding with functional response." *Oikos* 121:569-578.

Taylor, Jason M., Jeffrey A. Back, Bryan W. Brooks, and Ryan S. King. 2016. "Consumer-mediated nutrient recycling is influenced by interactions between nutrient enrichment and the antimicrobial agent triclosan." *Freshwater Science* 35:856-872.

Urabe, J., M. Kyle, W. Makino, T. Yoshida, T. Andersen, and J. J. Elser. 2002. "Reduced light increases herbivore production due to stoichiometric effects of light/nutrient balance." *Ecology* 83:619-627.

Van de Waal, Dedmer B., Val H. Smith, Steven A. J. Declerck, Eva C. M. Stam, and James J. Elser. 2014. "Stoichiometric regulation of phytoplankton toxins." *Ecology letters* 17:736-742.

Van de Waal, Dedmer B., Jolanda M. H. Verspagen, Miquel Lürling, Ellen Van Donk, Petra M. Visser, and Jef Huisman. 2009. "The ecological stoichiometry of toxins produced by harmful cyanobacteria: an experimental test of the carbon-nutrient balance hypothesis." *Ecology letters* 12:1326-1335.

Van den Brink, Paul J., Alistair B. A. Boxall, Lorraine Maltby, Bryan W. Brooks, Murray A. Rudd, Thomas Backhaus, David Spurgeon, Violaine Verougstraete, Charmaine Ajao, Gerald T. Ankley, and others. 2018. "Toward sustainable environmental quality: Priority research questions for Europe." *Environmental toxicology and chemistry* 37:2281-2295.

Vandenberg, Laura N., Theo Colborn, Tyrone B. Hayes, Jerrold J. Heindel, David R. Jacobs Jr, Duk-Hee Lee, Toshi Shioda, Ana M. Soto, Frederick S. Saal, Wade V. Welshons, and others. 2012. "Hormones and endocrine-disrupting chemicals: low-dose effects and nonmonotonic dose responses." *Endocrine reviews* 33:378-455.

Villar-Argaiz, M, JM Medina-Sánchez, and P Carrillo. 2002. "Linking life history strategies and ontogeny in crustacean zooplankton: implications for homeostasis." *Ecology* 7:1899-1914.

Villeneuve, D. L., D. Crump, N. Garcia-Reyero, M. Hecker, T. H. Hutchinson, C. A. LaLone, B. Landesmann, T. Lettieri, S. Munn, M. Nepelska, M. A. Ottinger, L. Vergauwen, and M. Whelan. 2014. "Adverse outcome pathway development II: best practices." *Toxicological sciences* 142(2):321–330.

Vrede, Tobias, Dean R. Dobberfuhl, S. A. L. M. Kooijman, and James J. Elser. 2004. "Fundamental connections among organism C: N: P stoichiometry, macromolecular composition, and growth." *Ecology* 85:1217-1229.

Wagner, Nicole D., Helmut Hillebrand, Alexander Wacker, and Paul C. Frost. 2013. "Nutritional indicators and their uses in ecology." *Ecology letters* 16:535-544.

Wagner, Nicole D., Brian P. Lankadurai, Myrna J. Simpson, Andre J. Simpson, and Paul C. Frost. 2014. "Metabolomic differentiation of nutritional stress in an aquatic invertebrate." *Physiological and Biochemical Zoology* 88:43-52.

Walker, Colin H., R. M. Sibly, and David B. Peakall. 2016. *Principles of ecotoxicology*. CRC press.

Wang, Wen-Xiong and Robert C. H. Dei. 2006. "Metal stoichiometry in predicting Cd and Cu toxicity to a freshwater green alga *Chlamydomonas reinhardtii*." *Environmental Pollution* 142:303-312.

Wanger, Nicole D., Felicia S. Osburn, Jingyu Wang, Raegyn B. Tylor, Asklynn R. Boedecker, Kevin C. Chambliss, Bryan Brooks, and Thad J. Scott. 2019. "Biological stoichiometry regulates toxin production in Microcystis aeruginosa (UTEX 2385)." *Toxins* 11(601).

Wang, W.-X., Nicholas S. Fisher, and Samuel N. Luoma. 1996. "Kinetic determinations of trace element bioaccumulation in the mussel *Mytilus edulis*." *Marine Ecology Progress Series* 140:91-113.

Wang, Hao, Yang Kuang, and Irakli Loladze. 2008. "Dynamics of a mechanistically derived stoichiometric producer-grazer model." *Journal of Biological Dynamics* 2:286-296.

Wang, Lin, Qixing Zhou, Lingling Ding, and Yuebing Sun. 2008. "Effect of cadmium toxicity on nitrogen metabolism in leaves of *Solanum nigrum L.* as a newly found cadmium hyperaccumulator." *Journal of hazardous materials* 154:818-825.

Weller, Florian, Lee-Anne Cecchini, Lynne Shannon, Richard B. Sherley, Robert J. M. Crawford, Res Altwegg, Leanne Scott, Theodor Stewart, and Astrid Jarre. 2014. "A system dynamics approach to modelling multiple drivers of the African penguin population on Robben Island, South Africa." *Ecological Modelling* 277:38-56.

Worm, Boris, Heike K. Lotze, Helmut Hillebrand, and Ulrich Sommer. 2002. "Consumer versus resource control of species diversity and ecosystem functioning." *Nature* 417(6891):848-851.

Xie, Congbo, Meng Fan, and Wei Zhao. 2010. "Dynamics of a discrete stoichiometric two predators one prey model." *Journal of Biological Systems* 18:649-667.

Yamamichi, Masato, Cédric L. Meunier, Angela Peace, Clay Prater, and Megan A. Rúa. 2015. "Rapid evolution of a consumer stoichiometric trait destabilizes consumer-producer dynamics." *Oikos* 124:960-969.

STOICHIOMETRIC ECOTOXICOLOGY
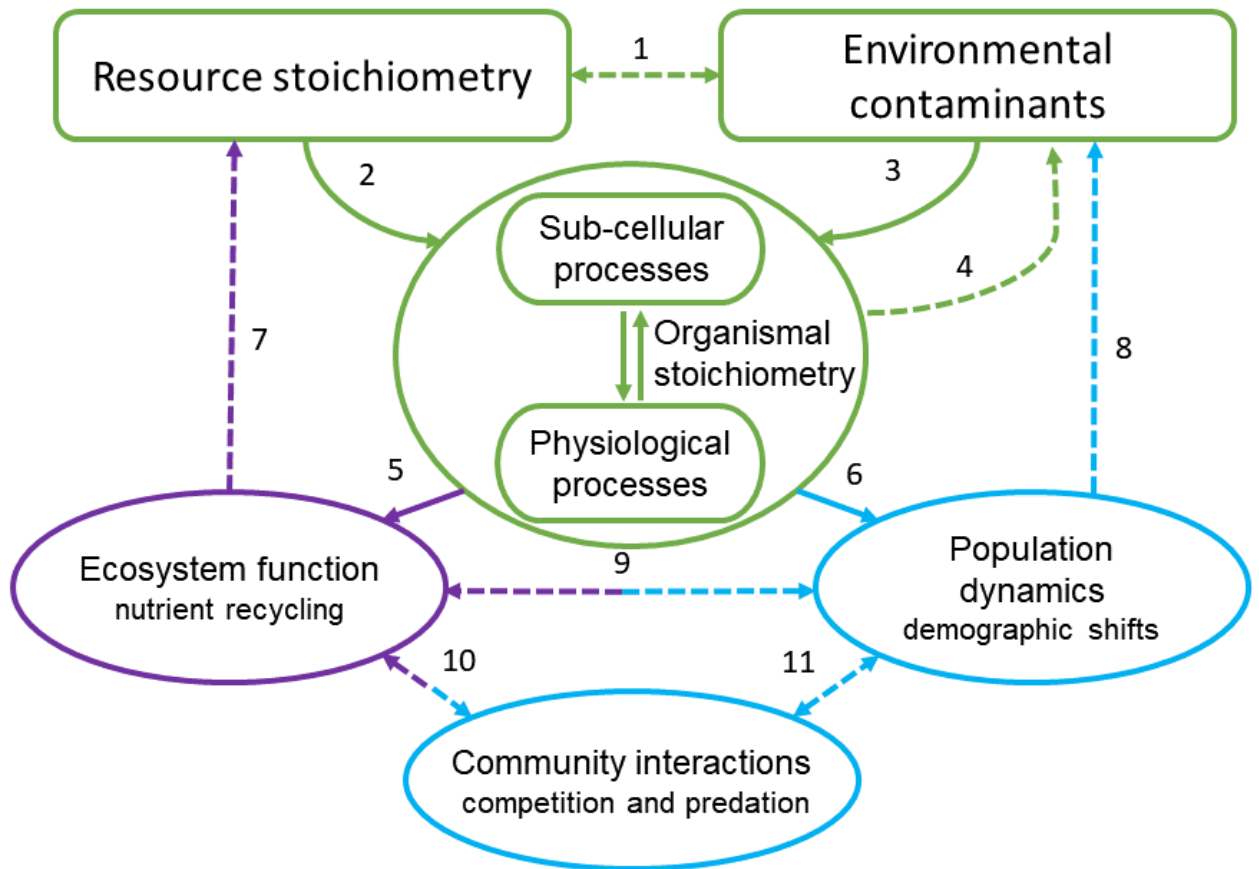
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Figure 1. Possible interactions of resource stoichiometry (i.e., nutrient ratios in the
environment) and environmental contaminants on relationships from the sub-organismal
level through ecosystem level. These effects include feedbacks within and between levels
of organization. Colors represent three categories of interaction: Organismal and sub-
organismal (green), population and community (blue), and ecosystem level
stoichiometric ecotoxicology (purple). Dashed arrows represent indirect mechanisms
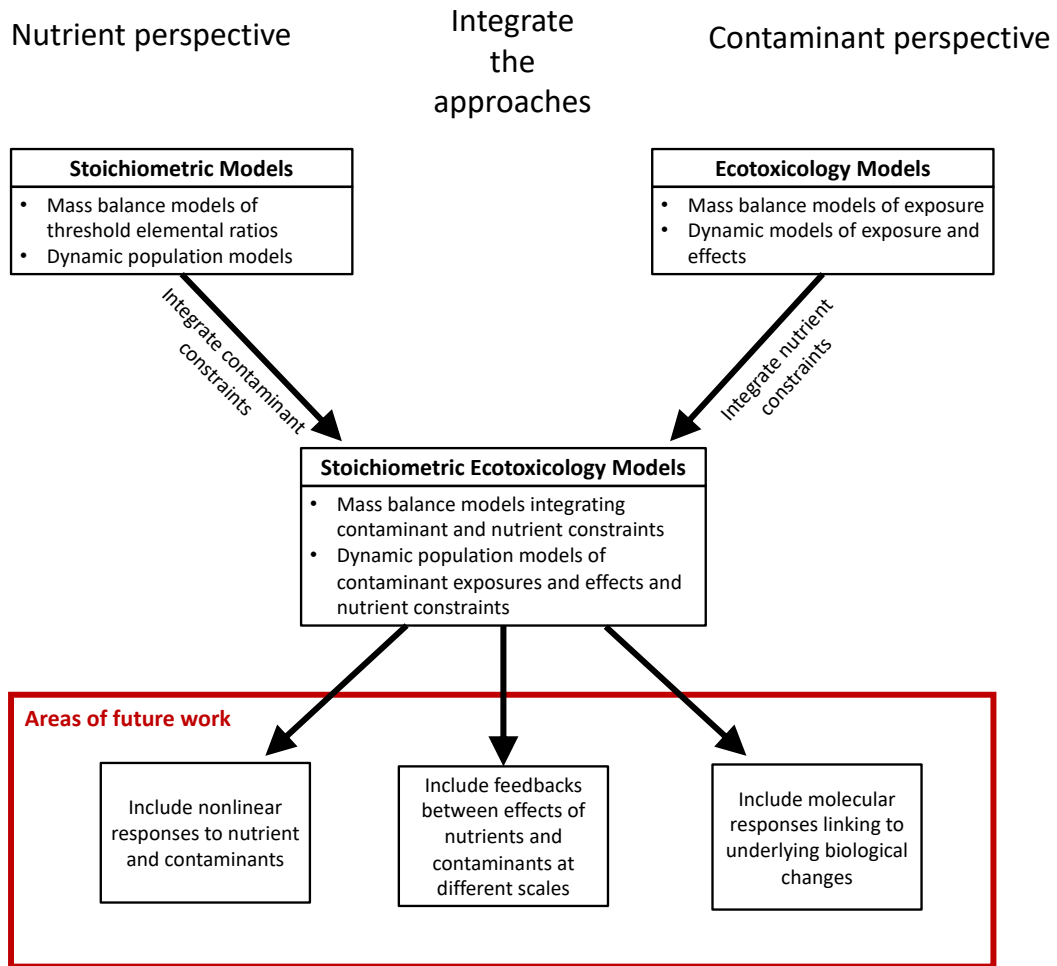and/or indirect feedbacks among components. Arrows are numbered and explained in the
accompanying text.

1
2 **Figure 2.** Proposed integrative modeling frameworks have promising potential to
3 explicitly incorporate nutrients and contaminants, as well as, integrative effects between
4 them. Stoichiometric Ecotoxicology models can then be extended to incorporate and
5 guide areas of future work listed in the red box.
6

Table 1. Examples of ecotoxicology studies that have examined stoichiometric mechanisms.

Effect	Contaminant	Biological model / ecological process	Stoichiometric parameter	Study scale	Type of ecosystem	Type of approach	Reference
Effects of the contaminant on dissolved nutrient availability and stoichiometry							
	Aluminium	n.a.	Mineral nutrient stoichiometry	Ecosystem	Freshwater	Observational	Kopacek et al. 2000
	Aluminium	Microbial decomposers	Mineral nutrient stoichiometry	Community	Freshwater	Microcosms	Clivot et al. 2014
Effects of basal resources stoichiometry on contaminant bioavailability in the medium							
	Silver nanoparticles	Zooplankton	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Conine & Frost 2017
Effects of the contaminant on basal resources C:N:P ratios)							
	Triclosan	Plant	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Fulton et al. 2010
	Atrazine	Periphyton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	(Murdock and Wetzel 2
	Silver	Microbial decomposers	Plant detritus C:N:P ratios	Community	Freshwater	Microcosms	Arce Funck et al. 2013
	Silver nanoparticles	Phytoplankton	Primary producers C:N:P ratios	Community	Freshwater	Microcosms	Das et al. 2014.
	Titanium nanoparticles	Cyanobacteria	Primary producers C:N:P ratios	Community	Freshwater	Microcosms	Cherchi et al. 2015
	Isoproturon, Mesosulfuron	Plant	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Nuttens et al. 2016
	Chlorpyrifos	Insect	Animal C:N:P ratios	Individual	Freshwater	Microcosms	Janssens et al. 2017
Effects of resources elemental composition on consumers tolerance to contaminants							
	Fluoxetine	Zooplankton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Hansen et al. 2008
	Triclosan	Plant	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Fulton et al. 2010.
	Glyphosate	Zooplankton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Lessard & Frost 2012
	Uranium, Arsenic	Plant	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Mkandawire & Dudel 2
	Silver	Plant	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Bian et al. 2013
	Imidachloprid	Zooplankton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Ieromina et al. 2014
	Metolachlor	Plant	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Brooks et al. 2015
	Atrazine	Phytoplankton	Mineral nutrient stoichiometry	Population	Freshwater	Microcosms	Baxter et al. 2016
	Silver	Macroinvertebrate	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Arce Funck et al. 2016
	Silver nanoparticles	Zooplankton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Conine & Frost 2017
	Cadmium	Macroinvertebrate	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Arce Funck et al. 2018

Effects of resources stoichiometry on consumers bioaccumulation of the contaminant

Contaminant	Organism	Stoichiometry	Level	Environment	Approach	Reference
Cadmium, Copper	Phytoplankton	Mineral nutrient stoichiometry	Population	Freshwater	Microcosms	Wang & Dei 2006
Mercury	Zooplankton	Primary producers C:N:P ratios	Individual	Freshwater	Microcosms	Karimi et al. 2007
n.a.	n.a.	Primary producers C:N:P ratios	n.a.	Freshwater	Theoretical model	Peace et al. 2016
Cadmium	Macroinvertebrate	Plant detritus C:N:P ratios	Individual	Freshwater	Microcosms	Arce Funck et al. 2018

Towards the use of stoichiometric approaches including non essential, potentially toxic elements

Contaminant	Organism	Stoichiometry	Level	Environment	Approach	Reference
Diverse metals	Macroinvertebrates	Primary producers Multielemental ratios		Freshwater	Observational	Karimi & Folt 2006
Cadmium, Copper	Phytoplankton	Mineral nutrient stoichiometry	Population	Freshwater	Microcosms	Wang & Dei 2006

Interactive effects of contaminants and stoichiometry on ecosystem functions/processes

Contaminant	Organism	Stoichiometry	Level	Environment	Approach	Reference
Zinc	Leaf litter microbial decomposition	Mineral nutrient stoichiometry	Community	Freshwater	Microcosms	Fernandes et al. 2009
Silver	Leaf litter microbial decomposition	Mineral nutrient stoichiometry	Community	Freshwater	Microcosms	Arce Funck et al. 2013
Atrazine	Periphyton primary production	Mineral nutrient stoichiometry	Community	Freshwater	microscosm	(Murdock, Shields Jr., a Lizotter Jr. 2013)
Silver nanoparticles	Phytoplankton primary production	Mineral nutrient stoichiometry	Community	Freshwater	Microcosms	Das et al. 2014
Silver	litter decomposition by macroinvertebrate	Plant detritus C:N:P ratios	Individual	Freshwater	Microcosms	Arce Funck et al. 2016
Cadmium	litter decomposition by macroinvertebrate	Plant detritus C:N:P ratios	Individual	Freshwater	Microcosms	Arce Funck et al. 2018

Effects of nutrient stoichiometry on the production of toxic compounds by organisms

Contaminant	Organism	Stoichiometry	Level	Environment	Approach	Reference
Plant secondary compounds	Plant	Mineral nutrient stoichiometry	Individual	Terrestrial	Literature review	Bryant et al. 1983
Cyanotoxins	Cyanobacteria	Mineral nutrient stoichiometry	Population	Freshwater	Microcosms	Van de Waal et al. 2009
Plant secondary compounds	Plant	Mineral nutrient stoichiometry	Individual	Terrestrial	Field experiment	Rivas-Ubach et al. 2012
Cyanotoxins	Phytoplankton/Cyanobacteria	Mineral nutrient stoichiometry	n.a.	Freshwater	Literature review	Van de Waal et al. 2014
Cyanotoxins	Cyanobacteria	Mineral nutrient stoichiometry	Population	Freshwater	Micrososm	(Wanger et al. 2019)
Plant secondary compounds	Plant	Mineral nutrient stoichiometry	Individual	Freshwater	Microcosms	Nuttens et al. 2016