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Pim de Voogt *Editor*

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Foreword

International concern in scientific, industrial, and governmental communities over traces of xenobiotics in foods and in both abiotic and biotic environments has justified the present triumvirate of specialized publications in this field: comprehensive reviews, rapidly published research papers and progress reports, and archival documentations These three international publications are integrated and scheduled to provide the coherency essential for nonduplicative and current progress in a field as dynamic and complex as environmental contamination and toxicology. This series is reserved exclusively for the diversified literature on "toxic" chemicals in our food, our feeds, our homes, recreational and working surroundings, our domestic animals, our wildlife, and ourselves. Tremendous efforts worldwide have been mobilized to evaluate the nature, presence, magnitude, fate, and toxicology of the chemicals loosed upon the Earth. Among the sequelae of this broad new emphasis is an undeniable need for an articulated set of authoritative publications, where one can find the latest important world literature produced by these emerging areas of science together with documentation of pertinent ancillary legislation.

Research directors and legislative or administrative advisers do not have the time to scan the escalating number of technical publications that may contain articles important to current responsibility. Rather, these individuals need the background provided by detailed reviews and the assurance that the latest information is made available to them, all with minimal literature searching. Similarly, the scientist assigned or attracted to a new problem is required to glean all literature pertinent to the task, to publish new developments or important new experimental details quickly, to inform others of findings that might alter their own efforts, and eventually to publish all his/her supporting data and conclusions for archival purposes.

In the fields of environmental contamination and toxicology, the sum of these concerns and responsibilities is decisively addressed by the uniform, encompassing, and timely publication format of the Springer triumvirate:

Reviews of Environmental Contamination and Toxicology [Vol. 1 through 97 (1962–1986) as Residue Reviews] for detailed review articles concerned with any aspects of chemical contaminants, including pesticides, in the total environment with toxicological considerations and consequences.

Bulletin of Environmental Contamination and Toxicology (Vol. 1 in 1966) for rapid publication of short reports of significant advances and discoveries in the fields of air, soil, water, and food contamination and pollution as well as methodology and other disciplines concerned with the introduction, presence, and effects of toxicants in the total environment.

Archives of Environmental Contamination and Toxicology (Vol. 1 in 1973) for important complete articles emphasizing and describing original experimental or theoretical research work pertaining to the scientific aspects of chemical contaminants in the environment.

The individual editors of these three publications comprise the joint Coordinating Board of Editors with referral within the board of manuscripts submitted to one publication but deemed by major emphasis or length more suitable for one of the others.

Coordinating Board of Editors

Preface

The role of *Reviews* is to publish detailed scientific review articles on all aspects of environmental contamination and associated (eco)toxicological consequences. Such articles facilitate the often complex task of accessing and interpreting cogent scientific data within the confines of one or more closely related research fields.

In the 50+ years since *Reviews of Environmental Contamination and Toxicology* (formerly *Residue Reviews)* was first published, the number, scope, and complexity of environmental pollution incidents have grown unabated. During this entire period, the emphasis has been on publishing articles that address the presence and toxicity of environmental contaminants. New research is published each year on a myriad of environmental pollution issues facing people worldwide. This fact, and the routine discovery and reporting of emerging contaminants and new environmental contamination cases, creates an increasingly important function for *Reviews.* The staggering volume of scientific literature demands remedy by which data can be synthesized and made available to readers in an abridged form. *Reviews* addresses this need and provides detailed reviews worldwide to key scientists and science or policy administrators, whether employed by government, universities, nongovernmental organizations, or the private sector.

There is a panoply of environmental issues and concerns on which many scientists have focused their research in past years. The scope of this list is quite broad, encompassing environmental events globally that affect marine and terrestrial ecosystems; biotic and abiotic environments; impacts on plants, humans, and wildlife; and pollutants, both chemical and radioactive; as well as the ravages of environmental disease in virtually all environmental media (soil, water, air). New or enhanced safety and environmental concerns have emerged in the last decade to be added to incidents covered by the media, studied by scientists, and addressed by governmental and private institutions. Among these are events so striking that they are creating a paradigm shift. Two in particular are at the center of ever increasing media as well as scientific attention: bioterrorism and global warming. Unfortunately, these very worrisome issues are now superimposed on the already extensive list of ongoing environmental challenges.

The ultimate role of publishing scientific environmental research is to enhance understanding of the environment in ways that allow the public to be better informed or, in other words, to enable the public to have access to sufficient information. Because the public gets most of its information on science and technology from internet, TV news, and reports, the role for scientists as interpreters and brokers of scientific information to the public will grow rather than diminish. Environmentalism is an important global political force, resulting in the emergence of multinational consortia to control pollution and the evolution of the environmental ethic. Will the new politics of the twenty-first century involve a consortium of technologists and environmentalists, or a progressive confrontation? These matters are of genuine concern to governmental agencies and legislative bodies around the world.

For those who make the decisions about how our planet is managed, there is an ongoing need for continual surveillance and intelligent controls to avoid endangering the environment, public health, and wildlife. Ensuring safety-in-use of the many chemicals involved in our highly industrialized culture is a dynamic challenge, because the old, established materials are continually being displaced by newly developed molecules more acceptable to federal and state regulatory agencies, public health officials, and environmentalists. New legislation that will deal in an appropriate manner with this challenge is currently in the making or has been implemented recently, such as the REACH legislation in Europe. These regulations demand scientifically sound and documented dossiers on new chemicals.

Reviews publishes synoptic articles designed to treat the presence, fate, and, if possible, the safety of xenobiotics in any segment of the environment. These reviews can be either general or specific, but properly lie in the domains of analytical chemistry and its methodology, biochemistry, human and animal medicine, legislation, pharmacology, physiology, (eco)toxicology, and regulation. Certain affairs in food technology concerned specifically with pesticide and other food-additive problems may also be appropriate.

Because manuscripts are published in the order in which they are received in final form, it may seem that some important aspects have been neglected at times. However, these apparent omissions are recognized, and pertinent manuscripts are likely in preparation or planned. The field is so very large and the interests in it are so varied that the editor and the editorial board earnestly solicit authors and suggestions of underrepresented topics to make this international book series yet more useful and worthwhile.

Justification for the preparation of any review for this book series is that it deals with some aspect of the many real problems arising from the presence of anthropogenic chemicals in our surroundings. Thus, manuscripts may encompass case studies from any country. Additionally, chemical contamination in any manner of air, water, soil, or plant or animal life is within these objectives and their scope.

Manuscripts are often contributed by invitation. However, nominations for new topics or topics in areas that are rapidly advancing are welcome. Preliminary communication with the Editor-in-Chief is recommended before volunteered review manuscripts are submitted. *Reviews* is registered in WebofScience™.

Inclusion in the Science Citation Index serves to encourage scientists in academia to contribute to the series. The impact factor in recent years has increased from 2.5 in 2009 to 7.0 in 2017. The Editor-in-Chief and the Editorial Board strive for a further increase of the journal impact factor by actively inviting authors to submit manuscripts.

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Feeding Behavioural Studies with Freshwater *Gammarus* spp.: The Importance of a Standardised Methodology

Giulia Consolandi, Alex T. Ford, and Michelle C. Bloor

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Abbreviation

AFDW Ash-free dry weight

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1 Introduction

Freshwater Gammarids are common leaf-shredding detritivores, and they usually feed on naturally conditioned organic material, in other words leaf litter that is characterised by an increased palatability, due to the action and presence of micro-organisms (Chaumot et al. 2015; Cummins 1974: Maltby et al. 2002). *Gammarus* spp. are biologically omnivorous organisms, so they are involved in shredding leaf litter and are also prone to cannibalism, predation behaviour (Kelly et al. 2002) and coprophagy when juveniles (McCahon and Pascoe 1988). *Gammarus* spp. is a keystone species (Woodward et al. 2008), and it plays an important role in the decomposition of organic matter (Alonso et al. 2009; Bundschuh et al. 2013) and is also a noteworthy prey for fish and birds (Andrén and Eriksson Wiklund 2013; Blarer and Burkhardt-Holm 2016). Gammarids are considered to be fairly sensitive to different contaminants (Ashauer et al. 2010; Bloor et al. 2005; Felten et al. 2008a; Lahive et al. 2015; Kunz et al. 2010); in fact Amphipods have been reported to be one of the most sensitive orders to metals and organic compounds (Wogram and Liess 2001), which makes them representative test organisms for ecotoxicological studies and valid sentinel species for assessing water quality status (Garcia-Galan et al. 2017).

Since Gammarids play an important role in the breakdown of organic matter in freshwater environments, it is understandable that their feeding behaviour is often used as a sublethal endpoint, to investigate water quality status and the effects of different contaminant types (Crane and Maltby 1991). Gammarid feeding activity could be altered by the presence of contaminants in the water, which could potentially alter their food source, influence the organism's biological function and cause abnormal behavioural responses. These types of feeding investigation have been carried out as in situ (i.e. directly in the environment) and ex situ (i.e. in the laboratory) studies (Bundschuh et al. 2011b; Dedourge-Geffard et al. 2009; Maltby et al. 2002; Zubrod et al. 2015). It has been demonstrated that feeding assays using Gammarids are representative of natural leaf decomposition in the environment (Maltby et al. 2002) and could be used to assess the effects of chemical contaminants and also understand the consequences of new-generation contaminants, such as plastic debris in freshwater environments (Blarer and Burkhardt-Holm 2016; Weber et al. 2018). Even though feeding behaviour studies have been carried out for almost half a century, there is a lack of standardisation for both ex situ and in situ methods. Without standardisation, there is a risk that the effects of a test substance could be under- or overestimated during in situ and ex situ approaches, which could reduce their usefulness in environmental biomonitoring programs. This paper aims to review the literature on feeding as an endpoint for amphipod ecotoxicology, by highlighting disparities in the published methodologies, and to help develop standardised protocols. Peer-reviewed literature was accessed through search engines, databases and library archives. In general, most feeding studies have reported four main stages: (1) acclimation period, (2) food preparation, (3) exposure and (4) end of the experiment and feeding rate calculation. The aforementioned four stages have been reviewed separately, and the variability of the published methodologies has been considered, in order to draw attention to the current discrepancies in the literature.

2 Acclimation Conditions

The first stage of an experiment (both in situ and ex situ) is the acclimation period that should be used to acclimate the organisms to the experimental conditions. However, the acclimation conditions are not always fully disclosed, and when they are, they sometimes contradict the experimental conditions. The reproducibility of an experiment is also highly dependent on many abiotic and biotic factors, which are rarely taken into consideration for Gammarid feeding studies (Coulaud et al. 2011). In the following sections, different variables (duration, temperature, light/dark cycles, type of water and organisms) that could impact the outcome of an experiment have been reviewed separately and summarised in Table 1, in order to emphasise the full range of variability within the literature. In some studies, Gammarids are sourced from laboratory breeding programs (e.g. Blockwell et al. 1996; Bloor and Banks 2006a, b; McCahon and Pascoe 1988).

2.1 Duration

Acclimation periods vary depending on the study (see Table 1), for example, Agatz et al. (2014) kept specimens of *Gammarus pulex* in the laboratory for 3 days prior to the start of the experiment, whereas another study left *Gammarus fossarum* organisms to acclimate for 21 days (Garcia-Galan et al. 2017). Typically the acclimation period used for Gammarids appears to be between 5 and 7 days, but some studies have selected longer intervals up to 35 days (see Table 1). Agatz and Brown (2014) stated that a 1-day acclimation period helped to reduce the variability of their results by just 1.6%, suggesting that a longer acclimation period could potentially have an even greater impact on reducing the intraspecific variability and consequently strengthen the statistics. Although experimental controls are incorporated into the majority of experimental designs, it becomes difficult to compare published peer-reviewed research when the test organisms have experienced anything between 3 and 35 days acclimation to laboratory conditions (Agatz et al. 2014; Garcia-Galan et al. 2017) (see Table 1), even more so when the organisms are used as water quality biomonitors for in situ experiments (see Table 1).

Table 1 Existing differences in the literature regarding *Gammarus* spp. acclimation conditions

Test organism	Age/sex/size organism	Duration	Temperature	Light/ dark cycle	Type of water Aeration pH	Type of study	References
Gammarus fossarum	Free from parasites No gravid females	7 days	16°C	12:12	Mixed water Aerated	Feeding and assimilation study	Blarer and Burkhardt-Holm (2016)
Gammarus fossarum	Adults with a cephalothorax length between 1.2 and 1.6 mm	7 days	15°C		River water	Feeding preferences study	Bundschuh et al. (2009)
Gammarus fossarum	Conducted as described by Bundschuh et al. (2009)					Feeding rate study	Bundschuh et al. (2011a)
Gammarus fossarum	Free from parasites Adults with a cephalothorax length between 1.2 and 1.6 mm	7 days	15°C		River water	Feeding rate study	Bundschuh et al. (2011b)
Gammarus fossarum	Free from parasites No gravid females Adults with a cephalothorax length between 1.2 and 1.6 mm	7 days	15°C		River water	Feeding rate study	Bundschuh et al. (2013)
Gammarus fossarum	Adults with a cephalothorax length between 1.2 and 1.6 mm	7 days	15°C	Total darkness	River and tap water mixture	Feeding behavioural study	Bundschuh et al. (2017)
Gammarus fossarum	Juveniles and adult males	15 days	12°C	10:14	Groundwater mixed with osmosed water Aerated	Ex situ and in situ feeding assay	Coulaud et al. (2011)
Gammarus fossarum	Dry mass = 6.8 ± 0.7 mg	7 days	10°C			Decomposition and feeding rate study	Danger et al. (2012)

Gammarus fossarum		20–25 days	12°C	10:14	Drilled groundwater Aerated	In situ feeding experiment	Dedourge-Geffard et al. (2009)
Gammarus fossarum	Adult males	21 days	12°C	10:14	Groundwater Aerated	Bioaccumulation study	Garcia-Galan et al. (2017)
Gammarus fossarum	Adult females	30–35 days	12°C	16:08	Drilled groundwater Aerated	Reproductive cycle and feeding study	Geffard et al. (2010)
Gammarus fossarum	Adult males with diameter from 1.6 to 2.0 mm	7 days	16°C	Total darkness	SAM-5S medium	Feeding rate study	Newton et al. (2018)
Gammarus fossarum		10 days	12°C	8:16	Drilled groundwater Aerated	Feeding behaviour and biomarkers analysis	Xuereb et al. (2009)
Gammarus fossarum	Free from parasites Adults with a cephalothorax length between 1.2 and 1.6 mm	7 days	15°C		River water Aerated	Feeding, accumulation and growth study	Zubrod et al. (2010)
Gammarus fossarum	Free from parasites Adult males (6–8 mm)	7 days	20°C	Total darkness	Aerated medium	Feeding and survival study	Zubrod et al. (2014)
Gammarus fossarum	Free from parasites Adult males (6–8 mm)	7 days	16°C	Total darkness	SAM-5S medium Aerated	Toxicity and feeding study	Zubrod et al. (2015)
Gammarus fossarum	Free from parasites Different sizes	3 days	16°C		SAM-5S medium Aerated	Feeding behavioural and physiological responses	Zubrod et al. (2017)
Gammarus pseudolimnaeus	Juveniles and adults					Feeding behavioural study	Bärlocher and Kendrick (1973b)
Gammarus pulex	Free from parasites Dry body mass 3.8–15 mg	3 days	13°C	12:12	Artificial pond water	Feeding rate study	Agatz et al. (2014)

(continued)

Table 1 (continued)

Test organism	Age/sex/size organism	Duration	Temperature	Light/dark cycle	Type of water Aeration pH	Type of study	References
Gammarus pulex	Organisms with parasites Both sexes Juveniles and adults	1 day	13°C	12:12	Artificial pond water Aerated pH = 7.4–7.9	Feeding rate studies	Agatz and Brown (2014)
Gammarus pulex	Free from parasites Adults (mean size 9.7 ± 1.4 mm) No gravid females	1. 4 days 2. 4 days	1. 15°C 2. 20°C		River water Artificial water Aerated	Feeding rate study with the Multispecies Freshwater Biomonitor	Alonso et al. (2009)
Gammarus pulex	3–7 mm		13°C	12:12	Dechlorinated tap water pH = 7.7	Feeding behavioural study	Blockwell et al. (1998)
Gammarus pulex	Adult males	7 days	15°C	12:12		In situ feeding assay	Crane and Maltby (1991)
Gammarus pulex	Males with first thoracic segment of 0.7–1.2 mm in size	7 days	15°C	12:12	River water	Feeding behavioural study	De Castro-Català et al. (2017)
Gammarus pulex	Adults (7–9 mm)	10 days	12°C		Well water pH = 7.19 ± 0.02	Physiological and behavioural responses	Felten et al. (2008a)
Gammarus pulex	Free from parasites Adult males (dry weight 6.5–12.0 mg)		15°C	12:12	Artificial pond water	In situ and laboratory feeding studies	Forrow and Maltby (2000)
Gammarus pulex	Adults (dry weight 8–10 mg)					Feeding behavioural study	Graça et al. (1993a)
Gammarus pulex	Adults (9–10 mm) Juveniles (2.5–3.5 mm)		15°C	12:12	Artificial pond water	Feeding behavioural study	Graça et al. (1993b)
Gammarus pulex	Adults		13°C			Feeding behavioural study	Hahn and Schulz (2007)

Gammarus pulex	Wet weight = 1.5–2.5 mg	10 days			Dechlorinated city tap water	Growth and feeding rate study	Hargeby and Petersen (1988)
Gammarus pulex	Both sexes		14°C	12:12	River water	Energetic state study	Iltis et al. (2017)
Gammarus pulex	Males (13–16 mm)		12°C	14:10	Aerated	Predation behaviour study	Kelly et al. (2002)
Gammarus pulex	Adults	7–14 days	19–22°C		Dechlorinated tap water Aerated pH = 8.28 ± 0.06	Feeding and bioaccumulation study	Lahive et al. (2015)
Gammarus pulex	Adult males (mean dry weight = 8.24 mg)	5–10 days	15°C	12:12	Artificial pond water	In situ feeding assay	Maltby et al. (2002)
Gammarus pulex	Adult males (dry weight = 7–10 mg)	7 days	15°C	12:12	Artificial pond water	Scope for growth assay	Naylor et al. (1989)
Gammarus pulex		1 day	14°C	16:8		Feeding behavioural study	Taylor et al. (1993)
Gammarus pulex	Adults Juveniles	7 days	16°C	16:8	ISO medium Aerated	Feeding activity and physiological responses	Weber et al. (2018)
Gammarus roeselii	Both sexes		15°C	12:12	Lake water	Feeding, assimilation and growth study	Gergs and Rothhaupt (2008)
Gammarus spp.		7 days			River water Aerated	Selective feeding study	Arsuffi and Suberkropp (1989)
Gammarus spp.		5 days	10°C		River water pH = 7.2	Physiological and behavioural responses	Maul et al. (2006)

2.2 Temperature

During the acclimation period, organisms need to be kept at a constant temperature and with a precise light/dark cycle. Gammarids from temperate countries are usually maintained at a temperature between 10 and 22°C (see Table 1). The temperature adopted in an experimental design is often selected to reproduce seasonal conditions, but unfortunately the literature does not always specify the selection criteria. Temperature can have a significant impact on Gammarids and on amphipods in general (Labaude et al. 2017). Foucreau et al. (2014) discovered that temperatures higher than 15°C altered various physiological parameters in *Gammarus pulex* populations in North France. Southern specimens consumed more oxygen at higher temperatures and had a higher glycogen content, which means they have a higher energy supply. Cold-acclimated organisms consumed more energy and oxygen when they are exposed to higher temperatures, and they presented a lower heat tolerance (Semsar-kazerouni and Verberk 2018). Interestingly, Alonso et al. (2009) acclimated their organisms at 15°C for 4 days, after which time the organisms were transferred to a 20°C room to acclimate for a further 4 days. Moving organisms from a low to a high temperature could have potentially affected the experimental results (Alonso et al. 2009). Furthermore, temperature plays an important role in the immune system of crustaceans (Le Moullac and Haffner 2000). Therefore, it is difficult to compare studies where the test animals have been acclimated at different temperatures, as this could have influenced their energy stores or their immune systems, for example. These differences could also be reflected in the organisms' behavioural reactions, which could be incorrectly interpreted as a result of exposure to specific contaminants. In fact, both Nilsson (1974) and Coulaud et al. (2011) reported an increased feeding rate with an increased temperature. The extent of the feeding rate increase was also dependent on leaf species (i.e. *Alnus glutinosa* or *Fagus sylvatica*) (Nilsson 1974). Acclimation temperature plays an even greater role in in situ experiments where the chosen temperature should be as close as possible to real-life environmental conditions. Interestingly, Coulaud et al. (2011) linked temperature and feeding rate through a linear regression, in order to better understand the impact of temperature on the Gammarids feeding. It was found that a small increase in mean temperature (from 12 to 13°C) could enhance the feeding rate by 7.3%.

2.3 Light and Dark Cycles

The same principle could be applied to the different light/dark cycles used during the acclimation period. The most commonly adopted light/dark cycle is 12:12 h (see Table 1) that reflects typical equinox conditions. However, some studies acclimate their organisms in total darkness, and in other studies, the adopted cycle is not specified (see Table 1). Sometimes a seasonal cycle is selected, in order to replicate the time of year when the organisms are collected from the wild, such as summer with a light/dark cycle of 16:8 h (Weber et al. 2018) or autumn with a cycle of 10:14 h (Garcia-Galan et al. 2017) (see Table 1). Adopting different light/dark cycles could make the comparison between studies challenging, since light could influence the organisms' physiological processes and behaviour (Perrot-Minnot et al. 2013).

2.4 Media Selection

The type of media selected for an experiment is another factor that could have an impact on the outcome of a study. Some researchers prefer to use an artificial medium (see Table 1) that guarantees standardisation (Agatz et al. 2014; Maltby et al. 2002), and in other studies, river water is sometimes used as a medium. However, river water might be contaminated, and this could therefore interfere with the organisms' cleansing process during their acclimation period, which makes it a peculiar choice of test media. Numerous studies have also used river water or a mixture (Alonso et al. 2009; Blarer and Burkhardt-Holm 2016; Bundschuh et al. 2009, 2017; De Castro-Català et al. 2017; Dedourge-Geffard et al. 2009; Gergs and Rothhaupt 2008; Iltis et al. 2017; Maul et al. 2006; Zubrod et al. 2015) (see Table 1). For example, Bundschuh et al. (2017) combined river water with tap water, which also has limitations as the tap water could be contaminated (Magi et al. 2018). Potentially, any type of water could be contaminated, which is why the authors recommend that researchers should report the chemical breakdown (i.e. presence of contaminants) of their chosen water media along with their study findings so that any contamination is transparent.

Gammarus pulex allocates up to 11% of its energy supply to osmotic regulation (Sutcliffe 1984), and Gammarids have been proven to be acid-sensitive (*Gammarus fossarum*; Felten and Guerold 2001; *Gammarus pulex*, Sutcliffe and Carrick 1973). In fact, acidic conditions induce a range of physiological and behavioural alterations, such as a reduction in the ventilation activity of *Gammarus pulex* (Felten et al. 2008b). These findings highlight the importance of measuring pH, as a shift in pH might influence the outcome of an experiment and prevent comparisons between studies. pH is rarely reported and presumably not measured in the environment during the collection process, the acclimation period or the experiment. Along with the chemical parameters of the acclimation media, the authors also recommend that pH is another factor that should be measured during the acclimation period, to ensure that accurate baseline data is recorded.

2.5 Characteristics of the Test Organism

Another important factor that plays a fundamental role in the reproducibility of a feeding experiment is the organism itself. Organisms of different age and sex may behave or respond differently to contaminants. For example, juveniles are more