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Review

Does zebra mussel (*Dreissena polymorpha*) represent the freshwater counterpart of *Mytilus* in ecotoxicological studies? A critical review

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ABSTRACT

One of the fundamentals in the ecotoxicological studies is the need of data comparison, which can be easily reached with the help of a standardized biological model. In this context, any biological model has been still proposed for the biomonitoring and risk evaluation of freshwaters until now. The aim of this review is to illustrate the ecotoxicological studies carried out with the zebra mussel *Dreissena polymorpha* in order to suggest this bivalve species as possible reference organism for inland waters. In detail, we showed its application in biomonitoring, as well as for the evaluation of adverse effects induced by several pollutants, using both *in vitro* and *in vivo* experiments. We discussed the advantages by the use of *D. polymorpha* for ecotoxicological studies, but also the possible limitations due to its invasive nature.

1. Introduction

The application of the European Water Framework Directive (WFD; Directive 2000/60/EC) for the surveillance of chemical contamination of surface waters involves two main objectives based on the assessment of the chemical status of water bodies, by determining whether contamination levels are compliant with the regulatory Environmental Quality Standards (EQSs), and the evaluation of temporal trends of the contamination in the different environmental compartments of aquatic ecosystems (Besse et al., 2012). Although to reach the first objective, checking compliance with EQSs, the chemicals' analyses in water are a requirement for all the priority substances, biota is starting to be a pivotal matrix as demonstrated by the addition of three biota EQSs (mercury, hexachlorobenzene and hexachlorobutadiene) in the Directive 2008/105/EC. The same Directive promotes the use of the so-called “integrating matrices”, such as biota and sediments, to reach the second objective of the WFD, contamination trend biomonitoring. In particular, biota is recognized as a preferential matrix for 14 and an optional matrix for 12 of the 41 substances listed under Directive 2008/105/EC. Furthermore, the physic-chemical characteristics of the 15 newly proposed substances also point to biota as a relevant matrix for 12 of these (Besse et al., 2012). In this context, it is crucial the selection of the proper biological model, which should possess several fundamental characteristics. Looking at the choice of organisms, aquatic macroinvertebrates emerge as one of the most valuable option, as they enable robust control of biotic factors, by using size homogenous indicator species that lend themselves well to practical and easy-to-handle biological models. Moreover,

Abbreviations: MTT, 3(4,5-dimethyl-2thiazholyl)-2,5-diphenyl-2H-tetrazolium bromide); MDMA, 3,4-methylenedioxymethamphetamine; AChE, acetylcholinesterase; ABM, active biomonitoring; ALP, alkaline phosphatase; AP, alkylphenols; Al, aluminum; Am, americium; As, arsenic; AhR, aryl hydrocarbon receptor; ATL, atenolol; Ba, barium; Bq, becquerel; B[α]P, benzo[α]pyrene; BE, benzoylecgonine; 2DE, bidimensional electrophoresis; BLM, bleomycin; BTs, butyltins; Cd, cadmium; Ca, calcium; CBZ, carbamazepine; CAT, catalase; Ce, cerium; Cs, cesium; ChEs, cholinesterases; Cr, chromium; Co, cobalt; Cu, copper; COES, crude organic extract of sediments; Cox I, cytochrome c oxidase subunit – I; DBT, dibutyltin; DDTs, dichlorodiphenyltrichloroethane and relative homologues; DCF, diclofenac; DMAA, dimethylarsinic acid; *dl*-PCBs, dioxin-like polychlorinated biphenyls; EDs, endocrine disruptors; ENDO, endosulphan; EROD, ethoxyresorufin-O-deethylase; EST, expressed sequence tag; HHCB, galaxolide; GEM, gemfibrozil; GSH, glutathione; GPx, glutathione peroxidase; GR, glutathione reductase; GST, glutathione-*S*-transferase; Hsp70, heath shock protein 70; HCB, hexachlorobenzene; HCHs, hexachlorocyclohexanes; I, iodine; LDH, lactate dehydrogenase; Pb, lead; LPO, lipid peroxidation; Mg, magnesium; Hg, mercury; MT, metallothionein; MeHg, methylmercury; MN, micronuclei; MMC, mitomycin C; MOA, mode (mechanism) of action; MBT, monobutyltin; MXR, multixenobiotic resistance; NRRA, neutral red retention assay; Ni, nickel; Nb, niobium; NSAIDs, non steroidal anti-inflammatory drugs; OCs, organochlorine compounds; PBM, passive biological monitoring; PMF, peptide mass fingerprint; POPs, persistent organic pollutants; PCPs, personal care products; P-gp, pglycoprotein; PPCPs, pharmaceutical and personal care products; PBDEs, polybrominated diphenyl ethers; PCBs, polychlorinated biphenyls; PCDDs, polychlorinated dibenzodioxins; PCDFs, polychlorinated dibenzofurans; PAHs, polycyclic aromatic hydrocarbons; K, potassium; PCR, potassium chromate; PCC, protein carbonyl content; PP2A, protein phosphatase 2A; Rb, rubidium; Se, selenium; Ag, silver; SCGE assay, single cell gel electrophoresis assay; SB, strand breaks; Sr, strontium; SOD, superoxide dismutase; Th, thorium; Sn, tin; AHTN, tonalide; TBT, tributyltin; TCS, triclosan; TPT, triphenyltin; U, uranium; VTG, vitellogenin-like proteins; Zn, zink; Zr, zirconium.

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invertebrates represent about 95% of animal species, have an important ecological role and could be potential transfer of pollutants through the food web (Baun et al., 2008). For all these reasons, mussels are widely used as sentinel organisms to monitor chemical pollution in the aquatic environment. In fact, mussels are filter feeding, sessile bottom dwellers that bioaccumulate many contaminants with little metabolic transformation and provide time-integrated observations of chemical contamination in the environment (Roesijadi et al., 1984; Sericano, 1993). For instance, the US NOAA (United States National Oceanic and Atmospheric Administration) Mussel Watch Program is the nation's longest running continuous coastal contaminant monitoring program. In particular, different *Mytilus* species (*Mytilus edulis*, *Mytilus californianus*, *Mytilus galloprovincialis*, *Mytilus trossulus*) and oysters (*Cressostrea virginica*) have been used to evaluate the contamination status of the US coasts. On the other hand, specimens from the genus *Mytilus* are historically used as sentinel organisms worldwide to monitor the contamination of some persistent organic pollutants (Monirith et al., 2003; Carro et al., 2005; Kožul et al., 2011), heavy metals and organotin compounds (Chase et al., 2001; Furdek et al., 2012; Edwards et al., 2014), radionuclides (Rožmarić et al., 2013; Kiliç et al., 2014) and more recently also emerging pollutants, such as pharmaceuticals (McEneff et al., 2014). *Mytilus* is used also to evaluate the effect of pollutants on biota, which represents the second part of the environmental risk assessment (Forbes and Forbes, 1994). Several studies have been conducted with different approach, such as classical bioassays (Beiras and Bellas, 2008; Marcheselli et al., 2010; Paredes et al., 2014), biomarkers (Canesi et al., 2010; Gonzalez-Rey and Bebianno, 2014; Franzellitti et al., 2014) genomics (Ioannou et al., 2009; Canesi et al., 2014; Liu et al., 2014) and proteomics (Schmidt et al., 2014; Hu et al., 2014). Although *Mytilus* is the most used biological model for the monitoring of coastal waters and for the effect evaluation of marine pollutants, no specific organism has been proposed for freshwaters. Thus, one of the future crucial challenges in the biomonitoring and ecotoxicological studies on the inland water bodies should be the identification of suitable organisms with convenient characteristics like *Mytilus*. Actually, a very similar biological model is present in the European and United States freshwaters, namely the invasive mollusc zebra mussel (*Dreissena polymorpha*; Pallas, 1771). This bivalve is a relatively widespread macrobenthic species in both lentic and lotic waters and is able to live in estuarine environments because it tolerates salinity up to 5‰. It is unique among the freshwater bivalves because, like marine ones, it releases gametes freely into the surrounding waters (Neumann and Jenner, 1992). Its ecological success is due mostly to its ability to develop through several larval stages (trochophora, veliconcha, pediveliger) and to attach itself by the bissus to hard substrates in both lakes and river beds. These ecological characteristics point out a phylogenetic affinity with *Mytilus* narrower than that with the native freshwater bivalves, such as *Unionids*. This is indirectly supported by studies of Stepien et al. (1999) and Orlova et al. (2005), in which *M. edulis* was used as the out group to evaluate phylogenetic relationships among dreissenids.

The following characteristics make the zebra mussel particularly useful as bioindicator: wide distribution, continuous availability throughout the year, adequate body size, sessile organism, ease of sampling, high longevity (3–5 years; Stanczykowska, 1977), great resistance at laboratory conditions. Moreover, zebra mussel has an enormous filtering capacity, ranging from 5 to 400 mL/bivalve/h (Baldwin et al., 2002) that allows a fast intake of environmental pollutants. This is a great advantage for the biomonitoring purposes because this sentinel-organism responds very quickly to contamination changes. Furthermore, the rapid intake of toxicants allows

Table 1
List of many biomonitoring studies using *D. polymorpha*. See the Glossary for the abbreviations' meaning.

Study area	Chemical	Range	Reference
Mosel River, France	^{110m}Ag	<d.l.–6 Bq/kg d.w.	Mersch et al., 1992
	^{58}Co	<d.l.–160 Bq/kg d.w.	
	^{60}Co	<d.l.–6 Bq/kg d.w.	
	^{134}Cs	<d.l.–2 Bq/kg d.w.	
	^{137}Cs	6–11 Bq/kg d.w.	
	^{40}K	25–54 Bq/kg d.w.	
	^{210}Pb	<d.l.–30 Bq/kg d.w.	
	^{232}Th	25–40 Bq/kg d.w.	
	^{238}U	6–24 Bq/kg d.w.	
Kiev Reservoir, Ukraine	^{140}Ba	<d.l.–2294 Bq/kg	Frantsevich et al., 1996
	^{141}Ce	<d.l.–2183 Bq/kg	
	^{144}Ce	28–1036 Bq/kg	
	^{134}Cs	11–666 Bq/kg	
	^{131}I	17–370 Bq/kg	
	^{103}Ru	<d.l.–2220 Bq/kg	
	^{106}Ru	<d.l.–740 Bq/kg	
	^{90}Sr	662–1273 Bq/kg	
	^{95}Zr, Nb	<d.l.–3219 Bq/kg	
Mirgenbach Reservoir, France	Cd	0.5–1.7 μg/g	Mersch et al., 1996a
	Cu	32.0–238.0 μg/g	
Buffalo, New York	Ag	<0.04 mg/kg w.w.	Roper et al., 1996
	As	0.97 mg/kg w.w.	
	Ba	7.00 mg/kg w.w.	
	Cd	0.47 mg/kg w.w.	
	Hg	0.03 mg/kg w.w.	
	Pb	3.28 mg/kg w.w.	
	Se	0.90 mg/kg w.w.	
	PAHs	6.58 mg/kg w.w.	
	PCB Aroclor 1248	1.64 mg/kg w.w.	
Lake Erie, Canada–United States	HCBs	19.1–41.3 μg/kg lip. w.	Roe and MacIsaac, 1998
	PCBs	5367.3–10465.7 μg/kg lip. w.	
Vienna, Austria	Cd	0.50–1.30 μg/g d.w.	Gundacker, 1999
	Cu	6.9–12.3 μg/g d.w.	
	Pb	0.18–1.11 μg/g d.w.	
	Zn	98.0–128.0 μg/g d.w.	
Lake Maggiore, Italy	DDTs	18.5–134.1 ng/g d.w.	Binelli et al., 2001a,b
Subalpine Lakes, Italy	Cd	0.60–3.44 μg/g d.w.	Camusso et al., 2001
	Co	0.88–1.51 μg/g d.w.	
	Cr	2.03–4.97 μg/g d.w.	
	Cu	14.6–26.3 μg/g d.w.	
	Hg	0.049–0.158 μg/g d.w.	
	Ni	11.9–24.2 μg/g d.w.	
	Pb	1.96–5.87 μg/g d.w.	
	Zn	158.00–346.00 μg/g d.w.	
St. Lawrence River, Canada–United States	DBT	<1–158 Sn/g w.w	Regoli et al., 2001
	MBT	<1–134 Sn/g w.w	
	TBT	37–1078 ng Sn/g w.w.	
	TPT	<1–52 ng Sn/g w.w.	
Buffalo, New York	DBT	0.47–2.25 ng Sn/g d.w.	Roper et al., 2001
	MBT	<1.97 ng Sn/gd.w.	
	TBT	3.50–5.88 ng Sn/g d.w.	

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