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Life cycle toxicity assessment of earthworms exposed to cadmium-contaminated soils

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Abstract Cadmium (Cd) is of great concern in the soil environment and it can damage terrestrial organisms. The purpose of this study was to employ a toxicokinetic/toxicodynamic (TK/TD) approach to investigate the effects of toxicologically relevant Cd accumulation on the life cycle growth of earthworms (*Lumbricus rubellus* and *Eisenia fetida*) and to assess potential terrestrial ecosystem risk. We reanalyzed growth toxicity and whole body and pellet accumulation data linked with TK/TD and life cycle growth models to estimate key rate constants. The growth risk of earthworms exposed to Cd was also assessed. This study found that the estimated whole body killing rate constant ($0.114\ g\ d\ \mu g^{-1}$) was much lower than that of pellet ($0.248\ g\ d\ \mu g^{-1}$). The recovery rate constant for whole body ($6.02\ d^{-1}$) was much higher than that of pellet ($2.91\ d^{-1}$). We also employed a life cycle-based probabilistic risk assessment model to estimate the growth inhibition risk for earthworms in response to environmentally relevant concentrations of Cd in Taiwan. Results showed that earthworms had a 90% growth inhibition probability risk of body weight, which was lower than 872.33 mg based on assessment of toxicologically relevant Cd accumulation. This study suggests that toxicologically relevant Cd accumulation could accurately reflect the capacity of Cd toxicity to earthworms. The integrated life cycle toxicity of earthworms exposed to Cd in this study provides a robust and applicable tool for the management of ecological risk assessment of Cd-contaminated soil.

Keywords Cadmium · Earthworm · Life cycle toxicity · Agricultural soils · Ecological risk assessment

Introduction

An increasing body of evidence from both observations and model simulations indicates that metal contamination of soil has altered terrestrial ecosystems locally and globally (Wang and Qin 2007; Wuana and Okieimen 2011). Anthropogenic sources of cadmium (Cd) emission include intensive agricultural activity (Cd used as pesticides and fertilizers) and industrial waste (Menzie et al. 2009). Cd is a non-essential element and is non-biodegradable. This can lead to accumulation and a decline in soil quality. It is a potential hazard for food safety and has adverse effects on environmental health. In particular, cadmium contamination in agricultural soils and its accumulation in crops and subsequent transfer to food chains is a global environmental concern (Rizwan et al. 2016).

Terrestrial invertebrates contact contaminated soil leading to accumulation of metal in cells and tissues. Earthworms are key bioindicators for monitoring soil ecosystem health and ecotoxicological risk because they are widely distributed and are less mobile. Thus, they are constantly exposed to soil contaminants especially for *Lumbricus rubellus* and *Eisenia fetida* (Paoletti 1999; Pirooznia et al. 2007; Haeba et al. 2013). Previous studies indicated that earthworms could accumulate high levels of metals in the

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soil environment (Morgan and Morgan 1988; Nahmani et al. 2009). Moreover, earthworms can easily concentrate metals that are more likely to transfer to higher trophic levels (Bohlen 2016). *E. fetida* is widely used in soil ecological studies and is also used as a fish bait in Taiwan (Blakemore et al. 2006).

Seeking effective approaches for assessing the Cd-soil environmental risk remains a challenge. Previous studies used body residues of Cd to relate biologically toxic responses (Stürzenbaum et al. 2004; Nakashima et al. 2008; Veltman et al. 2008). However, some studies indicated that Cd bound specific subcellular species such as metallothionein, which then made it unavailable. This resulted in different potential biological consequences of whole-tissue Cd accumulation (Dubois and Hare 2009; Li et al. 2009a). Li et al. (2009a) also found that most Cd accumulated as a toxicologically unavailable fraction in *E. fetida* and only 3% of Cd was found in the organelles. Hence, using the toxicologically relevant Cd accumulation as the pellet fraction of tissue (heat sensitive protein or organelles) might provide adequate and applicable means to assess Cd toxicity to organisms.

To assess environmental risk, full life cycle toxicity is an effective approach to evaluate the potential hazards of metals to organisms (Agrawal and Gopal 2013). Environmentally relevant exposure concentrations are often lower than that of laboratory concentrations. It is difficult to observe chemical caused physiological effects in organism during short-term periods. Therefore, the effective endpoints of toxicity such as growth and the development of organisms are more important in long-term exposure. West et al. (2001) developed an ontogenetic growth model that could describe the body biomass trajectory of organisms from birth to maturity based on the properties of optimized energy distribution networks. The energy metabolic allocation is not balanced between supply and demand. This ultimately limits growth—especially when organisms are exposed to contaminant stressors. The integration of an ontogenetic growth model and dynamic energy budget (DEB) theory can appropriately assess the risk of growth dynamics in life cycle events under arsenic exposure (Tsai and Liao 2006).

USEPA (2007) suggested that metal risk assessment framework should include a toxicokinetic/toxicodynamic (TK/TD) scheme to better understand the ability of organisms in regulating and accumulating metals. Therefore, well-known metal exposure routes and metal TKs should be considered with respect to metal toxicity prediction and ecological health risk assessment. This study employed a TK/TD approach to investigate the effects of toxicologically relevant Cd accumulation on the life cycle growth toxicity of earthworms (*L. rubellus* and *E. fetida*) and to examine whether toxicologically relevant Cd accumulation can be a candidate for assessing terrestrial ecosystem risk. To gain insight into how to predict the effective life cycle toxicity risk of earthworms in Cd-contaminated soil environments, we hypothesized the following: (i) the pellet fraction is more likely to be the toxicologically relevant biomarker of toxic effects, (ii) Cd accumulation is a toxicologically relevant biomarker and is more related to toxic effects, (iii) integrating Cd accumulation in toxicologically relevant biomarkers and growth toxicity into the TK/TD model can estimate more sensitive parameters, and (iv) the integrated model can better assess ecotoxicological life cycle risk in earthworms from Cd contaminated soil.

Materials and methods

Study data

The valuable database was provided by Spurgeon et al. (2004). We examined soil Cd toxicity to earthworms based on growth biomass responses. The experiments used five Cd exposure concentrations (0, 12.5, 50, 200, and 800 $\mu g\,L^{-1}$) to monitor the growth biomass of juvenile earthworms (*L. rubellus*) during 294 days of exposure (Spurgeon et al. 2004). The experiments provided a potential growth toxicity assessment to understand Cd toxicity.

For Cd accumulation, Conder et al. (2002) conducted an exposure bioassay by using the earthworm *E. fetida* exposed to 1574 $mg\,kg^{-1}$ Cd for 14 days. Therefore, data adopted from Conder et al. (2002) were reanalyzed to estimate the physiological determinants such as uptake and elimination rate constants from the whole body and pellet. The pellet fraction is more likely to be the toxicologically-relevant biomarker of toxic effects.

This study adopted growth toxicity and Cd accumulation data to develop the integrated model. Growth toxicity data of *L. rubellus* were adopted to estimate specific growth coefficients via the growth toxicity model (toxicodynamics). This used Cd accumulation data of *E. fetida* to estimate toxicokinetic parameters via the bioaccumulation model. We then incorporated experimental data from both species with estimated parameters to estimate toxicodynamic parameters (damage and recover levels) of Cd stress via a damage assessment model.

Bioaccumulation model

A first-order biokinetic model was used to fit the Cd accumulation data to estimate the uptake rate constant k_1 ($g\,g^{-1}\,d^{-1}$) and elimination rate constant k_2 (d^{-1}). The

following bioaccumulation model was used,

$$C_b(t) = C_b(0)e^{-k_2 t} + \frac{k_1}{k_2} C_s\left(1 - e^{-k_2 t}\right), \quad (1)$$

Here, $C_b(t)$ is the Cd body burden varied with time t (mg kg^{-1} wet wt), $C_b(0)$ is the initial Cd concentration of body (mg kg^{-1} wet wt), and C_s is the Cd concentration in soil (mg kg^{-1}).

The bioaccumulation factor (BAF) was determined from the equation: $BAF = k_1/k_2$. To obtain uptake rate and elimination rate constants for pellets from the subcellular fraction, a bioaccumulation model was used to fit the selected data.

Damage assessment model

The relationship between external Cd concentration and growth inhibition of earthworm was used to represent the dose-response profile. The damage assessment model (DAM) was used to estimate the killing rate (k_k) and recovery rate (k_r) constants. The DAM includes three dynamic variables: (i) the cumulative damage ($D(t)$), (ii) the cumulative hazard ($H(t)$), and (iii) the susceptibility probability ($S(t)$).

First, damage accumulation and damage recovery were used to describe the $D(t)$ coupling bioaccumulation model (Eq. (1)) (Lee et al. 2002),

$$dD(t)/dt = k_a C_b(t) - k_r D(t), \quad (2)$$

Then $D(t)$ can be solved as,

$$D(t) = k_a \frac{k_1}{k_2} C_s \left(\frac{e^{-k_r t} - e^{-k_2 t}}{k_r - k_2} + \frac{1 - e^{-k_r t}}{k_r} \right), \quad (3)$$

where $D(t)$ is the cumulative damage (−), k_a is the damage accumulation rate (g μg^{-1} d^{-1}), and k_r is the damage recovery rate constant (d^{-1}).

Second, the cumulative hazard ($H(t)$) was assumed to be proportional to $D(t)$. Thus, the crucial DAM parameter—the killing rate constant k_k (g μg^{-1} d^{-1})—could be determined by

$$H(t) = (k_k/k_a)D(t), \quad (4)$$

Finally, growth inhibition (%) as the physiological susceptibility probability ($S(t)$) was used to estimate important parameters, k_k and k_r, where ($S(t)$) can be derived directly from an exponential relationship of cumulative hazard (Lee et al. 2002; Ashauer et al. 2007),

$$S(t) = 1 - \exp(-H(t)) = 1 - \exp\left[-k_k \frac{k_1}{k_2} C_s \left(\frac{e^{-k_r t} - e^{-k_2 t}}{k_r - k_2} + \frac{1 - e^{-k_r t}}{k_r} \right) \right]. \quad (5)$$

Growth toxicity model

All organisms should fuel ontogenetic growth in their life. Energy is allocated to synthesize new biomass and to maintain existing biomass. The West growth model describes an organism's ontogenetic growth (biomass) trajectory from birth to maturity based on energy allocation without growth toxicity (West et al. 2001). This model is a quantitative model based on the energy consumption principle to describe the increasing growth biomass.

The West growth model has the following form:

$$W(t) = W_{\max,0} \cdot \left\{ 1 - \left[1 - \left(\frac{W_0}{W_{\max,0}} \right)^{1/4} \right] \exp\left(- \frac{A_0 t}{4 \cdot W_{\max,0}{}^{1/4}} \right) \right\}^4, \quad (6)$$

where $W(t)$ is the time-dependent body weight (g), W_0 is the body weight at birth of earthworm or approximately 3.95×10^{-7} g, $W_{\max,0}$ is the maximum body weight in an uncontaminated environment (2.11 g) (Spurgeon et al. 2004), and A_0 is a specific growth coefficient ($g^{1/4}$ d^{-1}) that could be estimated by optimal fitting Eq. (6) to the growth body weight profile.

Moreover, the toxicant dynamic energy budget (DEB_{tox}) describes the mode of action (MOA) of chemical toxicity that alters energy allocation (Kooijman and Bedaux 1996; Alunno-Bruscia et al. 2009). Hence, the integrated DEB_{tox} theory with the West growth model could simulate the metal toxicity on growth (biomass) inhibition. Tsai and Liao (2006) showed that the MOA of reducing food assimilation efficiency was well predicted by the West growth model (Tsai and Liao 2006), and thus Eq. (6) can be rewritten as

$$W(t) = \left[W_{\max,0}\left(1 - GI_{w/p}(t)\right) \right] \times \left\{ 1 - \left[1 - \left(\frac{W_0}{W_{\max,0}\left(1 - GI_{w/p}(t)\right)} \right)^{1/4} \right] \exp\left(- \frac{A_0 t}{4\left(W_{\max,0}\left(1 - GI_{w/p}(t)\right)\right)^{1/4}} \right) \right\}^4, \quad (7)$$

where $W_{\max 0}\left(1 - GI_{w/p}(t)\right)$ is the ultimate body weight of earthworm under the contaminated environment and $GI_{w/p}$ is the growth inhibition probability estimated from whole body or pellet bioaccumulation data.

Predictive growth inhibition risk model

To investigate relationships among earthworm body weight, growth inhibition, and soil Cd exposures, this study used a biologically-based DAM to reconstruct the dose-response

profile as,

$$GI_{w/p}(t) = 1 - \exp\left(-H_{w/p}(t)\right), \quad (8)$$

where $H_{w/p}(t)$ is the cumulative hazard level from soil Cd exposures based on whole body or pellet bioaccumulation data. The soil Cd contamination is located in southern Taiwan and was selected to implement the proposed model to estimate the growth inhibition risk.

The growth inhibition risk can be calculated as the probability density function (pdf) of southern Taiwan soil Cd concentrations combined with the conditional probability of growth inhibition ($GI_{w/p}$)-dependent body weight (W) (Eq. (7)) subjected to Cd concentration (C_s) as: $P(W(GI_{w/p})|C_s)$. Hence, the risk probability of growth inhibition for body weight could be expressed as,

$$P(R_W) = P(C_s) \times P\left(W\left(GI_{w/p}\right)|C_s\right), \quad (9)$$

where $P(R_W)$ is the probability risk for body weight inhibition and $P(C_s)$ is the pdf of external Cd concentration in soil. Furthermore, we selected various scenarios of Cd exposures including uncontaminated soil (0 μg g^{-1}), farmland soil (5 μg g^{-1}), and general soil (20 μg g^{-1}) standards in Taiwan (Taiwan EPA 2011) as representative concentrations. We also used the monitored value (51.37 μg g^{-1}) from soil in southern Taiwan to simulate the body weight dynamics of earthworms in response to Cd in the lifespan.

Uncertainty analysis and simulation scheme

TableCurve 2D (Version 5.0) (AISN Software Inc., Mapleton, OR, USA) packages were used to perform all curve fittings. The Crystal Ball® software (Version 2000.2, Decisioneering, Inc., Denver, Colorado, USA) was used to implement the Monte Carlo simulation to generate 2.5- and 97.5-percentiles as the 95% confidence interval (CI) for all fitted models. The risks of growth inhibition were generated from the cumulative distribution of simulation outcomes. The data showed that 10,000 interactions were sufficient to ensure the results. Figure 1 is the schematic illustration of this study for estimating the key parameters and representations of the mechanistic models.

Results

Biological responses of earthworm

Figure 2 depicts Cd accumulations in whole body and pellets of earthworms. These were best described by the bioaccumulation model ($r^2 = 0.83$–0.94). The rate constant estimates of uptake (k_1) and elimination (k_2) were 0.056 ± 0.008 g g^{-1} d^{-1} (mean ± SE) and 0.140 ± 0.036 d^{-1} and 0.032 ± 0.009 g g^{-1} d^{-1} and 0.369 ± 0.144 d^{-1}, for whole body and pellet, respectively. The rapid accumulation was found in whole body and pellet over the course of 14 days. The estimated BAFs of whole body and pellet were 0.40 and 0.09, respectively. However, both BAFs were less than 1.

We incorporated the TKs of whole body and pellet, respectively, into the DAM (Eq. 5). DAM was used to fit the constructed Cd-growth inhibition data of earthworm to estimate killing (k_k) and recovery (k_r) rate constants ($r^2 = 0.92$). The results indicated that estimated k_k of 0.114 g d μg^{-1} for whole body was much lower than that of 0.248 g d μg^{-1} for pellet, whereas the k_r of 6.02 d^{-1} for the whole body was much higher than that of 2.91 d^{-1} for pellet (Fig. 3).

Growth toxicity

From 12.5–800 μg g^{-1} Cd, obvious growth inhibition was observed in 200 and 800 μg g^{-1} Cd (Fig. 4d, e). Biomass losses even occurred in 50–800 μg g^{-1} Cd at 100 days (Fig. 4c, d, e). The best-fit of the reducing food assimilation efficiency demonstrates a biomass growth effect of earthworms under various Cd exposure scenarios with estimated model-specific growth cost coefficient A_0 ($r^2 = 0.87$–0.99) (Fig. 4, Table 1). The estimated growth cost coefficients ranged from 0.002–0.033 $g^{1/4}$ d^{-1} for Cd exposures, whereas the value for the control group was 0.03 ± 0.001 $g^{1/4}$ d^{-1}. These results indicated that the coefficients of growth costs depended on the Cd exposure concentrations and revealed that high concentration of Cd inhibited much more growth cost. However, 12.5 μg g^{-1} of Cd inhibited less growth than that of the control group (0 μg g^{-1}) (Table 1).

Risk assessment practices

The environmentally relevant soilborne Cd concentration in southern Taiwan had a lognormal (LN) distribution with a geometric mean (gm) of 42.23 μg g^{-1} and a geometric standard deviation (gsd) of 1.86 (Fig. 5a). The body weight can be calculated using the estimated soilborne Cd distribution as the input together with the TK/TD parameters (k_1, k_2, k_k, k_r, and A_0) used in the West growth model for the Cd-earthworm system. The results revealed that growth inhibition risk gradually increased as the body weight decreased for whole body and pellet (Fig. 5b, c).

Table 2 summarizes the threshold of body weight for probability of growth inhibition at risks of 0.1, 0.5, and 0.9 for the Cd-earthworm system. The results showed that earthworms had a 90% growth inhibition probability risk of

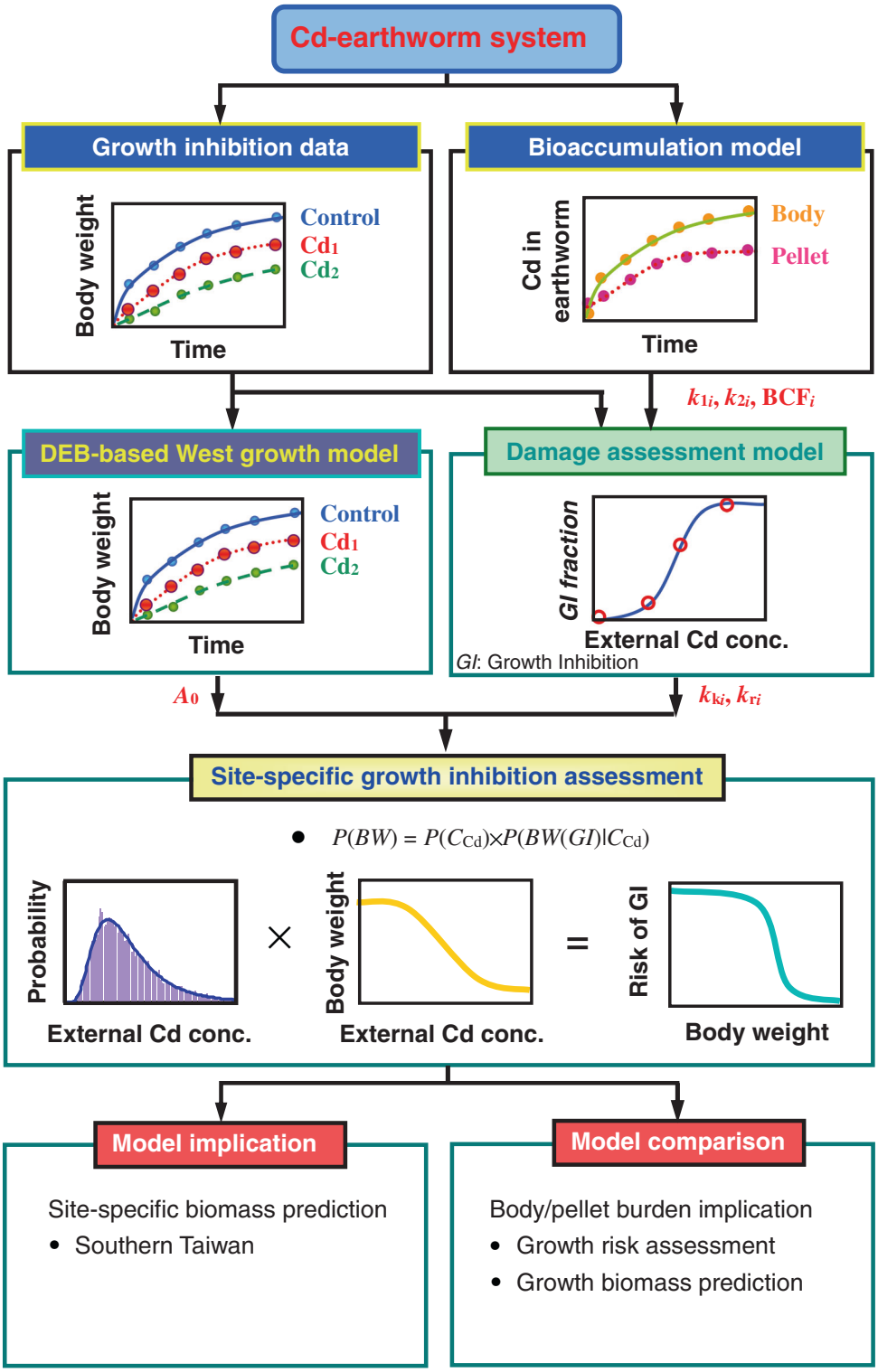

Fig. 1 Schematic presentation of growth inhibition risk assessment in earthworm exposed to cadmium via a linking bioaccumulation model, damage assessment model, and DEB-based West growth model (see text for detailed description of symbols)

body weight lower than nearly 873 mg for both whole body and pellet (Table 2).

We also adopted the control standards in farmland and general land soils in Taiwan (Taiwan EPA 2011) as well as the environmentally relevant soil Cd concentration in southern Taiwan (51.37 $\mu g\, g^{-1}$ (mean)) to simulate the dynamics of body weight changes in earthworms based on the biological parameters of whole body and pellet (Fig. 6). The results showed that the dynamics in body weight changed with respect to whole body and pellet; they appeared to have a similar pattern over time in response to various Cd levels in soil.

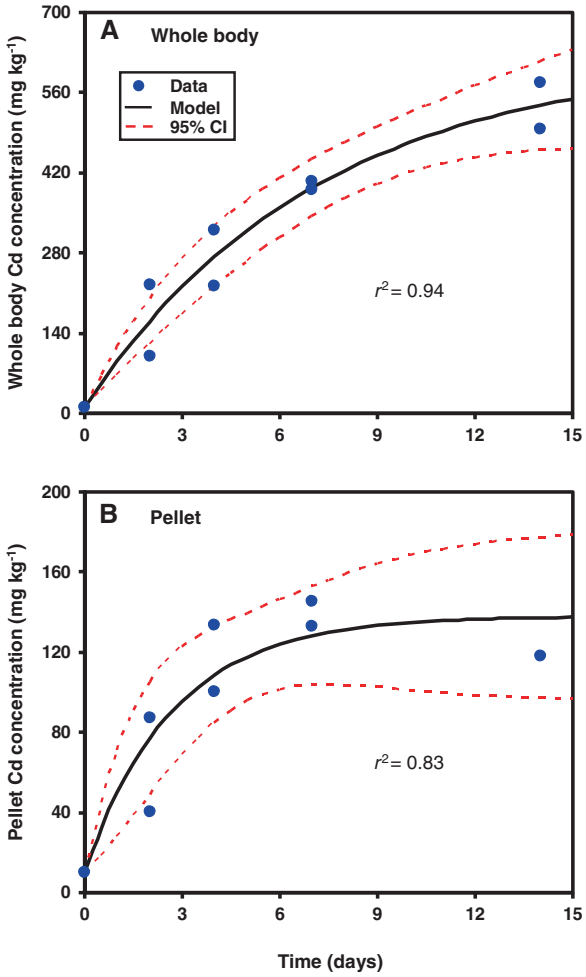

Fig. 2 **a** Whole body and **b** pellet Cd concentrations in earthworm exposed to 1574 mg kg^{-1} Cd for 14 days. Uptake rate (k_1) and elimination rate (k_2) constants were estimated by fitting data to bioaccumulation model. *Black solid* and *red dash line* represent the fitted model and 95% confidence intervals, respectively (colour figure online)

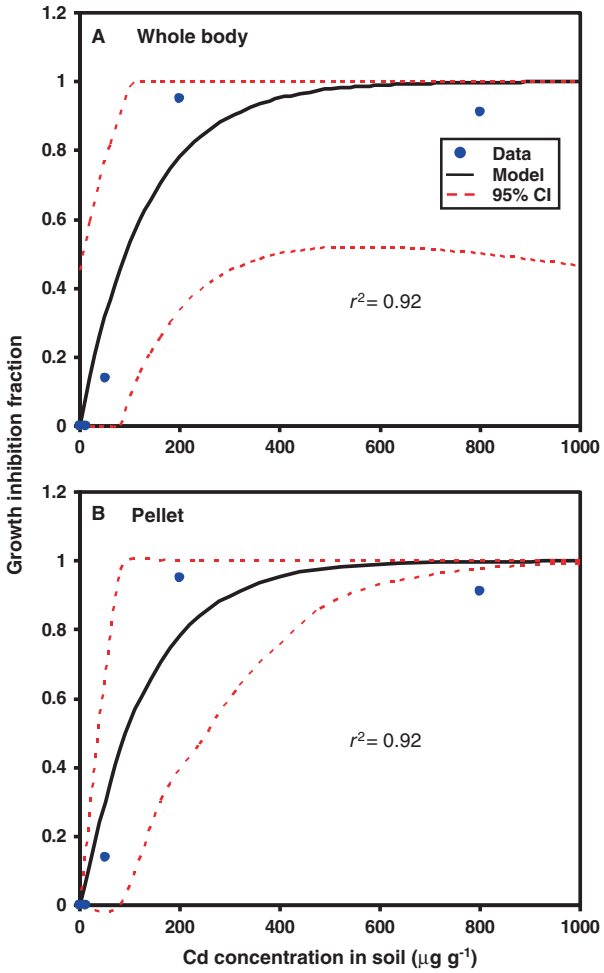

Fig. 3 Killing rate (k_k) and recovery rate (k_r) constants estimates for **a** whole body and **b** pellet by fitting the DAM-based safety function to the Cd concentrations in soil-course earthworm growth inhibition fraction profiles. *Black solid* and *red dash line* represent the fitted model and 95% confidence intervals, respectively (colour figure online)

Discussion

Toxicologically relevant Cd accumulation

This study estimated four toxicokinetic and toxicodynamic determinants (k_1, k_2, k_k, and k_r) based on the whole body and pellet accumulations to assess Cd growth inhibition risk and growth trajectories of earthworms. We found that the BAF (0.08) of Cd in whole body was much less than that of pellet (0.4). This study also found a strong influence of toxicokinetics with Cd distribution in the storage subcellular fraction. The results of estimated TK parameters agree with Li et al. (2009a) with respect to subcellular partitioning of earthworms in response to soil Cd contamination.

There are few studies on Cd accumulation in earthworms. This study adopted toxicokinetic and toxicodynamic data from different earthworm species (*E. fetida* and *L. rubellus*). Extrapolation between two species is possible with appropriate data and ecological principles. It has been shown that Cd induced MT-2 up-regulation was similar in *E. fetida* and *L. terrestris* (Asensio et al. 2007). A similar phylogenetic analysis of MT-2 nucleotide sequences was found in *L. terrestris* and *L. rubellus* (Gong et al. 2015). *E. fetida* and *L. rubellus* have similar Cd MT accumulation. Both have very low levels of MT expression in response to uncontaminated environment, but high MT accumulation and expression levels with Cd contamination were observed (Stürzenbaum et al. 1998; Gruber et al. 2000). Therefore, extrapolation between *E. fetida* and *L. rubellus* is plausible here.

We compared differences in Cd-induced growth risk of earthworms in southern Taiwan and found no differences in growth risks between TK/TD assessments based on whole body and pellet bioaccumulations. However, our results

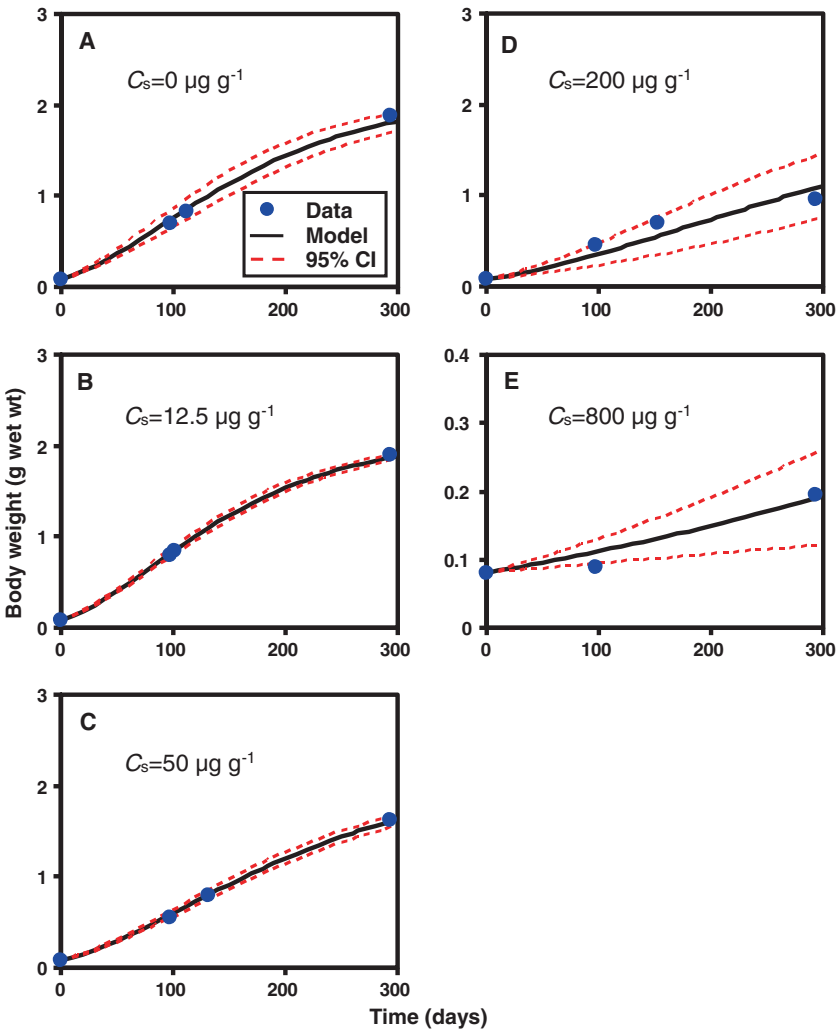

Fig. 4 Coefficients of growth costs (A_0) estimated via the DEB-based West growth model for different Cd exposure scenarios, **a** 0 g g^{-1} (control), **b** 12.5 g g^{-1}, **c** 50 g g^{-1}, **d** 200 g g^{-1}, and **e** 800 g g^{-1}. *Black solid* and *red dash line* represent the fitted model and 95% confidence intervals, respectively (colour figure online)

Table 1 Growth parameters used and estimated for earthworms exposed to cadmium

C_s (µg g^{-1})	0	12.5	50	200	800
W_0 (g)[a]	0.0807	0.0807	0.0807	0.0807	0.0807
W_{max} (g)[a]	2.11	2.11	2.11	2.11	2.11
A_0 (g$^{1/4}$ d^{-1})	0.030	0.033	0.024	0.015	0.002
r^2	0.99	0.99	0.99	0.87	0.93

[a] Adopted from Spurgeon et al. (2004)

indicated that the killing rate constant of pellet bioaccumulation was twice as high as whole body bioaccumulation. This indicates that using whole body accumulation to estimate the toxic effects will underestimate the capacity of Cd toxicity in earthworms. Furthermore, the recovery rate constant of pellet bioaccumulation was two-fold lower than whole body bioaccumulation. The estimated recovery capacity based on toxicologically relevant accumulation did not include the Cd influx to the biological detoxified fraction of tissue in which vulnerability and recovery of earthworms in the soil ecosystems might not be precisely evaluated. Hence, using only the site of Cd action to estimate k_k and k_r of growth inhibition was better than that of whole body bioaccumulation.

Most studies indicate that increased Cd bioaccumulation of earthworms depends on increasing Cd concentration and exposure time (Li et al. 2009a; Ibekwe et al. 2001; Ngu et al. 2006; Nahmani et al. 2007; Liang et al. 2009). However, Liang et al. (2011) and Ardestani et al. (2014) showed negative correlations between bioaccumulation factors of earthworms and increased Cd exposure indicating lower efficiency of Cd bioaccumulation when earthworms were exposed to higher Cd concentrations. This study estimated TK rate constants based on 1574 mg kg^{-1} constant Cd exposure to predict the bioaccumulation level and growth inhibition risks of earthworms in response to environmentally relevant concentrations of 0.75–150 mg

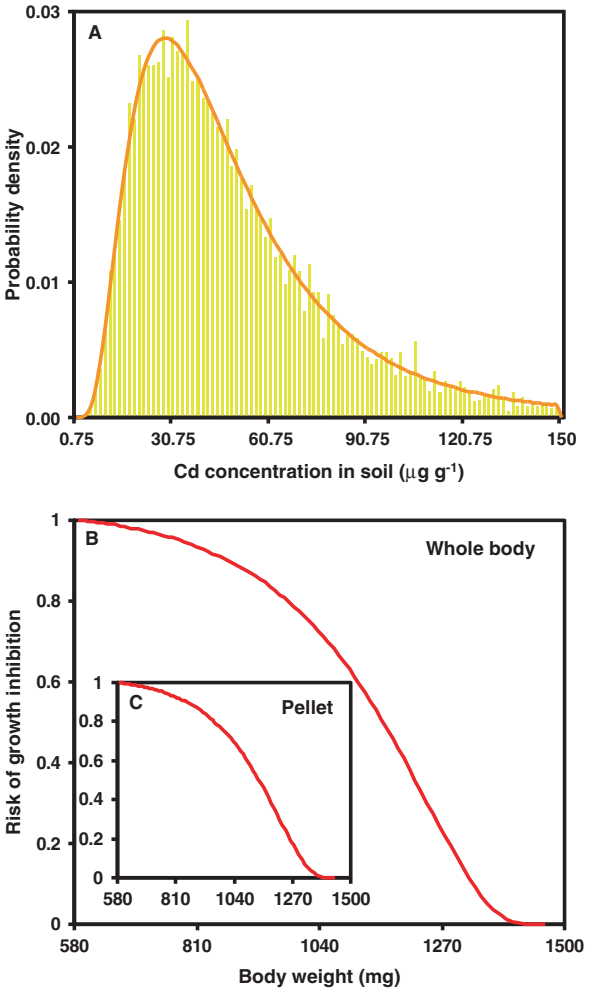

Fig. 5 **a** Probability density function of environmentally relevant Cd concentration in soil measured from southern Taiwan. The LN (42.23, 1.86) denotes a lognormal distribution with geometric mean of 42.23 μg g^{-1} and a geometric standard deviation of 1.86. Risk curves estimate the growth inhibition for earthworms exposed to environmentally relevant Cd concentrations over a lifespan of 300 days based on **b** whole body and **c** pellet input parameters

Table 2 Growth inhibition risk of earthworm based on whole body and pellet cadmium accumulation estimations

	Growth inhibition risk		
	0.1	0.5	0.9
	Body weight (mg)		
Whole body	1336.66	1160.13	873.14
Pellet	1336.40	1159.58	872.33

kg^{-1}. Thus, the soil ecological risk of Cd exposures was likely underestimated. This study suggests that Cd concentration-dependent TK parameters of toxicologically relevant bioaccumulation in earthworms provide applicable data that can be used in further risk assessment.

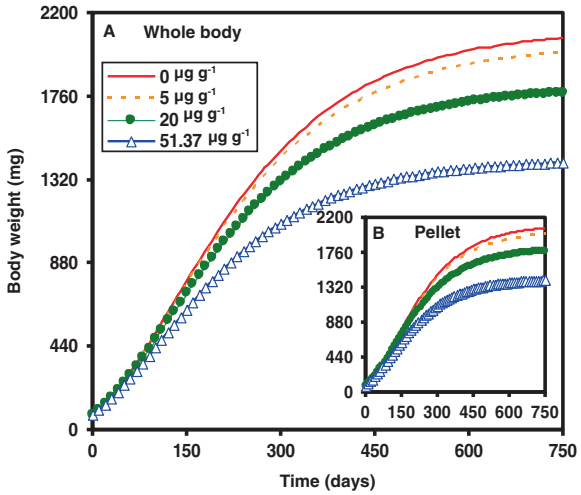

Fig. 6 Simulated body weights for earthworm exposed to 0 (control), 5, 20, and 51.37 g g^{-1} Cd in soil based on the input parameters of **a** whole body and **b** pellet. The amounts of 5 and 20 g g^{-1} Cd in soil are control standards in farmland- and general land-soils in Taiwan, respectively, whereas 51.37 g g^{-1} Cd in soil is the mean value measured in southern Taiwan

Effects of environmental properties on Cd toxicity

Cd toxicity of earthworms is influenced by the fraction of Cd available for uptake from the soil. Metal bioavailability is determined by environmental factors including soil physico-chemical properties, soil solution, and common soil cations. The highest toxicokinetic parameters of earthworms were related to cation exchange capacity and the uptake rate constant was higher at higher soil pH and lower cation exchange capacity (Peijnenburg et al. 1999; Giska et al. 2014). In the past decade, the competing relationship between cations (H related to pH) and Cd ions to bind to biotic ligands has been developed by biotic ligand models (BLM) with affinity determinants for assessing metal toxicity in the terrestrial environment (Li et al. 2008, 2009b, c). However, terrestrial BLM values for estimating Cd and cation affinity constants in earthworms are limited. The terrestrial BLM predicts Cd bioavailability and toxicity to a soil organism, and it is based on the effects of soil pore water composition. The soil solid phase should be considered when assessing metal bioavailability.

The biotic ligand model has been integrated with toxicokinetic models to offer a complete understanding of metal bioaccumulation, toxicity, and bioavailability in aquatic and soil environments (Liao et al. 2007; Veltman et al. 2010; Ardestani and van Gestel 2013). Metal bioaccumulation is influenced by the affinity and capacity of biotic ligands to bind free metal ions and competing cations or to complex with other organic ligands, rendering the metal poorly available to biotic ligand sites. Liao et al. (2007) indicated that the fraction of metal binding on biotic ligands

(bioavailability) may lead to metal transport influx in the fixed level of accumulation (toxicokinetics) and impair normal biological responses (toxicodynamics). Environmental factors, including organic matter content and pH, induced a stronger impact on earthworm life cycle parameters than soil Cd concentration (Nahmani et al. 2007). We suggest that integrating toxicologically relevant accumulation to interpret the biological effects of earthworms to soil environmental stressors. These might provide insights into the contribution of life cycle risk assessment in the soil environment.

Life cycle risk improvement

Previous studies showed that uptake and minimum elimination rate constants of chemical substances decreased with organism body weight (Hendriks et al. 2001; Hendriks and Heikens 2001). The Optimal Modeling for EcotoxicoloGical Applications (OMEGA) was developed by Hendriks et al. (2001) considering the effects of exposure concentration, trophic level and body weight of species on toxicokinetic assessment. The OMEGA model estimates these toxicokinetic parameters based on an allometric relation. This TK/TD assessment suggests that the TD rate constants were significantly influenced by toxicokinetic parameters. We suggest that the OMEGA model can incorporate the growth TD model to accurately estimate the Cd damage or hazard capacity on earthworm life cycle toxicity.

Earthworm life cycle parameters, including growth, maturation, and reproduction were significantly influenced by soil Cd (Kammenga et al. 1996). Field-contaminated Cd was the most crucial element to interpret differences in life cycle parameters of earthworms—it is highly associated with loss of body weight (Nahmani et al. 2007). A decreasing growth rate due to high soil Cd concentration resulted in a loss of body weight (Žaltauskaitė and Sodienė 2014).

This study used whole body and pellet Cd toxicokinetics and growth bioassay data of juvenile earthworms to assess the Cd-induced life cycle growth dynamics and risks of earthworms in Taiwan. The growth cost coefficient A_0 was estimated based on the limited information of growth toxicity and Cd accumulation at juvenile stages for earthworms exposed to soil Cd. Žaltauskaitė and Sodienė (2014) and found that the increased age of earthworms was associated with a decreased Cd-sensitive response. However, juvenile earthworms are more sensitive to Cd at an initial development stage. This can lead to serious consequences for population health with a significant impact on juveniles (Kammenga et al. 1996; Ramskov and Forbes 2008). The stage-specific growth parameters could also be incorporated into the life cycle growth model to provide a robust health assessment.

In conclusion, this study incorporated the toxicologically relevant Cd accumulation and growth cost coefficients to assess life cycle growth toxicity and risks of earthworm exposed to soil Cd. The TK/TD parameters based on Cd accumulation of pellet fraction is a more toxicologically relevant biomarker of Cd.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

Agrawal A, Gopal K (2013) Biomonitoring of water and waste water. Springer, New York

Alunno-Bruscia M, van der Veer HW, Kooijman SALM (2009) The AquaDEB project (phase I): analyzing the physiological flexibility of aquatic species and connecting physiological diversity to ecological and evolutionary processes by using dynamic energy budgets. J Sea Res 62:43–48

Ardestani MM, van Gestel CAM (2013) Using a toxicokinetics approach to explain the effect of soil pH on cadmium bioavailability to *Folsomia candida*. Environ Pollut 180:122–130

Ardestani MM, van Straalen NM, van Gestel CAM (2014) Uptake and elimination kinetics of metals in soil invertebrates: a review. Environ Pollut 193:277–295

Asensio V, Kille P, Morgan AJ, Soto M, Marigomez I (2007) Metallothionein expression and neutral uptake as biomarkers of metal exposure and effect in *Eisenia fetida* and *Lumbricus terrestris* exposed to Cd. Eur J Soil Sci Biol 43: S233–S238

Ashauer R, Boxall ABA, Brown CD (2007) New ecotoxicological model to simulate survival of aquatic invertebrates after exposure of fluctuating and sequential pulses of pesticides. Environ Sci Technol 41:1480–1486

Blakemore RJ, Chang CH, Chuang SC, Ito MT, James S, Chen JH (2006) Biodiversity of earthworms in Taiwan: a species checklist with confirmation and new records of exotic lumbricids *Eisenia fetida* and *Eiseniella tetraedra*. Taiwania 51:226–236

Bohlen PJ (2016) Earthworms. In: Lal R (ed) Encyclopedia of soil science, 3rd edn. CRC Press, New York

Conder JM, Seals LD, Lanno RP (2002) Methods for determing toxicologically relevant cadmium residues in the earthworm *Eisenia fetisa*. Chemosphere 4:1–7

Dubois M, Hare L (2009) Subcellular distribution of cadmium in two aquatic invertebrates: change over time and relationship to Cd assimilation and loss by predatory insect. Environ Sci Technol 43:356–361

Giska I, van Gestel CAM, Skip B, Laskowski R (2014) Toxicokinetics of metal in the earthworm *Lumbricus rubellus* exposed to natural polluted soils-relevance of laboratory tests to the field situation. Environ Pollut 190:123–132

Gong L, Li W, Li J, Li WE, Wu WR, Yu LW (2015) Cloning, characterization, and expression of cadmium-induced metallothionein-2 gene from earthworm *Pheretima aspergillum* (E. Perrier). Genet Mol Res 14:16782–16789

Gruber C, Stürzenbaum S, Gehrig P, Sack R, Hunziker P, Berger B, Dallinger R (2000) Isolation and characterization of a self-

sufficient on-domain protein (Cd)-metallothionein from *Eisenia foetida*. Eur J Biochem 267:573–582
Haeba M, Kuta J, Arhouma ZK, Elwerfalli HMA (2013) Earthworm as bioindicator of soil pollution around Benghazi City, Libya. J Environ Anal Toxicol 3:1000189
Hendriks AJ, Heikens A (2001) The power of size 2. Rate constants and equilibrium ratios for accumulation of organic substance related to octanol-water partition ratio and species weight. Environ Toxicol Chem 20:1421–1437
Hendriks AJ, van der Linde A, Cornelissen G, Sijm DTHM (2001) The power of size 1. Rate constants and equilibrium ratio and species weight. Environl Toxicol Chem 20:1399–1420
Ibekwe AM, Papiernik SK, Gan J, Yate SR, Yang CH, Crowley DE (2001) Impact of fumigants on soil microbial communities. Appl Environ Microbiol 67:3245–3257
Kammenga JE, Busschers M, Van Straalen NM, Jepson PC, Bakker J (1996) Stress induced fitness reduction is not determined by the most sensitive life-cycle trait. Funct Ecol 10:106–111
Kooijman SALM, Bedaux JJM (1996) The analysis of aquatic toxicity data. VU University Press, Amsterdam
Lee JH, Landrum PF, Koh CH (2002) Prediction of time-dependent PAH toxicity in *Hyalella Azteca* using a damage assessment model. Environ Sci Technol 36:3124–3130
Li L, Zhou D, Wang P, Peijnenburg WJGM (2009a) Kinetics of cadmium uptake and subcellular partitioning in the earthworm *Eisenia fetida* exposed to cadmium-contaminated soil. Arch Environ Contam Toxicol 57:718–724
Li LZ, Zhou DM, Luo XS, Wang P, Wang QY (2008) Effect of major cations and pH on the acute toxicity of cadmium to the earthworm *Eisenia fetida*: implications for the biotic ligand model approach. Arch Environ Contam Toxicol 55:70–77
Li LZ, Zhou DM, Wang P, Allen H, Sauvé S (2009c) Predicting Cd partitioning in spiked soils and bioaccumulation in the earthworm *Eisenia fetida*. Appl Soil Ecol 42:118–123
Li LZ, Zhou DM, Wang P, Jin SY, Peijnenburg WJGM, Rienecke AJ, van Gestel CAM (2009b) Effect of cation competition on cadmium uptake from solution by earthworm *Eisenia fetida*. Environ Toxicol Chem 28:1732–1738
Liang SH, Chen MH, Chen CC, Chen CS, Chen JH, Chen SC (2009) Impact of cadmium on bacterial communities in the gut of *Metaphire posthuma*. J Hazard Mater 172:1212–1217
Liang SH, Chen SC, Chen CY, Kao CM, Yang JI, Shieh BS, Chen JH, Chen CC (2011) Cadmium-induced earthworm metallothionein-2 is associated with metal accumulation and counteracts oxidative stress. Pedobiologia 54:333–340
Liao CM, Lin CM, Jou LJ, Chang KC (2007) Linking valve closure behavior and sodium transport mechanism in freshwater clam *Corbicula fluminea* in response to copper. Environ Pollut 147:656–667
Menzie CA, Ziccardi LM, Lowney YW, Fairbrother A, Shock SS, Tsuji JS, Hamai D, Proctor D, Henry E, Su SH, Kierski MW, McArdle ME, Yost LJ (2009) Importance of considering the framework principles in risk assessment for metals. Environ Sci Technol 43:8478–8482
Morgan JE, Morgan AJ (1988) Earthworms as biological monitors of cadmium, copper, lead and zinc in metalliferous soils. Environ Pollut 54:123–138
Nahmani J, Hodson ME, Black S (2007) Effects of metals on life cycle parameters of earthworm *Eisenia fetida* exposed to field-contaminated, metal-polluted soils. Environ Pollut 149:44–58
Nahmani J, Hodson ME, Devin S, Vijver MJ (2009) Uptake kinetics of metals by the earthworm *Eisenia fetida* exposed to field-contaminated soil. Environ Pollut 157:2622–2628
Nakashima T, Okada T, Asahi J, Yamashita A, Kawai K, kasai H, Matsuno K, Gamou S, Hirano T (2008) 8-Hydroxydeoxyguanosine generated in the earthworm *Eisenia fetida* grown in metal-containing soil. Mutat Res 654:138–144
Ngu TT, Sturzenbaum SR, Stilloman MJ (2006) Cadmium binding studies to the earthworm *Lumbricus rubellus* metallothionein by electrospray mass spectrometry and circular dichroism spectroscopy. Biochem Biophys Res Commun 351:229–233
Paoletti MG (1999) The role of earthworms for assessment of sustainability and as bioindicators. Agric Ecosyst Environ 74:137–155
Peijnenburg WJGM, Baerselman R, de Groot AC, Jager T, Posthuma L, Van Veen RPM (1999) Relating environmental availability to bioavailability: soiltype-dependent metal accumulation in the Oligochaete *Eisenia andrei*. Ecotoxicol Environ Safe 44: 294–310
Pirooznia M, Gong P, Guan X, Inouye LS, Yang K, Perkins EJ, Deng Y (2007) Cloning, analysis and functional annotation of expressed sequence tags from the earthworm *Eisenia fetida*. BMC Bioinform 8(Suppl. 7):S7
Ramskov T, Forbes VE (2008) Life history and population dynamics of the opportunistic polychaete *Capitella sp*. I in relation to sediment organic matter. Mar Ecol Prog Ser 69:181–192
Rizwan M, Ali S, Adrees M, Rizvi H, Zia-Ur-Rehman M, Hannan F, Qayyum MF, Hafeez F, Ok YS (2016) Cadmium stress in rice: toxic effects, tolerance mechanisms, and management: a critical review. Environ Sci Pollut Res Int 23:17859–17879
Spurgeon DJ, Svendsen C, Kille P, Morgan AJ, Weeks JM (2004) Responses of earthworm (*Lumbricus rubellus*) to copper and cadmium as determined by measurement of juvenile traits in a specifically designed test system. Ecotoxicol Environ Safe 57:54–64
Stürzenbaum SR, Kille P, Morgan AJ (1998) Heavy metal-induced molecular responses in the earthwarm *Lumbricus rubellus* genetic fingerprinting by directed differential display. Appl Soil Ecol 9:495–500
Stürzenbaum SR, Georgiev O, Morgan AJ, Kille P (2004) Cadmium detoxification in earthworms: from genes to cells. Environ Sci Technol 38:6283–6289
Taiwan EPA (2011) https://sgw.epa.gov.tw/public/07_Pollutant.asp?polluMethod=Soil
Tsai JW, Liao CM (2006) Mode of action and growth toxicity of arsenic to tilapia *Oreochromis mossambicus* can be determined bioenergetically. Arch Environ Contam Toxicol 50:144–152
US EPA (2007) Framework for metals risk assessment. US Environmental Protection Agency Office of the Science Advisor, Risk Assessment Forum
Veltman K, Huijbregts MA, Hendriks AJ (2008) Cadmium bioaccumulation factors for terrestrial species: application of the mechanistic bioaccumulation model OMEGA to explain field data. Sci Total Environ 406:413–418
Veltman K, Huijbregts MAJ, Hendriks AJ (2010) Integration of biotic ligand models (BLM) and bioaccumulation kinetics into a mechanistic framework for metal uptake in aquatic organisms. Environ Sci Technol 44:5022–5028
Wang XS, Qin Y (2007) Some characteristics of the distribution of heavy metals in urban topsoil of Xuzhou, China. Environ Geochem Health 29:11–19
West GB, Brown JH, Enquist BJ (2001) A general model for ontogenetic growth. Nature 413:628–631
Wuana RA, Okieimen FE (2011) Heavy metals in contaminated soil: a review of sources, chemistry, risks and best available strategies for remediation. ISRN Ecol 2011:402647
Žaltauskaitė J, Sodienė I (2014) Effects of cadmium and lead on the life-cycle parameters of juvenile earthworm *Eisenia fetida*. Ecotoxicol Environ Safe 103:9–16