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Assessment of selenium toxicity on the life cycle of *Caenorhabditis elegans*

Wen-Hsuan Li · Yun-Ru Ju · Chung-Min Liao · Vivian Hsiu-Chuan Liao

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Abstract Selenium (Se) is a growing problem of global concern. Se can cause adverse effects on reproductive systems, which have been linked to declines in animal populations. The soil nematode *Caenorhabditis elegans* (*C. elegans*) is a ubiquitous soil organism that is increasingly utilized as a model organism in aquatic and soil toxicology. In the present study, the experimental data for individual body length, survival rate, brood size, and hatching rate were used to evaluate the possible effects of selenite [Se(IV)] on *C. elegans*. A stage-classified matrix model was applied to the experimental data to provide information on the population dynamics of *C. elegans* and to assess the Se(IV)-affected asymptotic population growth rate. Estimates of the survival probability showed significant decreases in survival at all stages when *C. elegans* was exposed to Se(IV). The growth probability of *C. elegans* in the L1 stage showed the most significant decline, from 0.11 h^{-1} (for the control) to 0.04 h^{-1} [for exposure to 3 mM Se(IV)]. These results showed that Se(IV) has a profound impact on *C. elegans* population dynamics. The asymptotic population growth rate (λ) was found to range from 1.00 to 0.64 h^{-1} for increasing Se(IV) concentrations, implying a potential risk of population decrease for *C. elegans* exposure to a Se(IV)-contaminated environment. Our study shows how a mechanistic view based on the Se(IV) effects on the soil nematode *C. elegans* can promote a life cycle toxicity assessment. An important implication of this analysis is that mathematical models can be used to produce a population stage structure, to give clarity to the analysis of the key population-level endpoint (the asymptotic population growth rate) of population dynamics, and to evaluate the influences for the response of other species to environmental Se. These models sequentially provide candidate environmental criteria for the evaluation of the population impact of Se.

Keywords Selenium · *Caenorhabditis elegans* · Mathematical models · Population · Life cycle toxicity

Wen-Hsuan Li and Yun-Ru Ju have contributed equally to this work.

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W.-H. Li · Y.-R. Ju · C.-M. Liao · V. H.-C. Liao (✉)
Department of Bioenvironmental Systems Engineering, National Taiwan University, No. 1 Roosevelt Road, Sec. 4, Taipei 106, Taiwan
e-mail: vivianliao@ntu.edu.tw

Present Address:
Y.-R. Ju
Department of Bioenvironmental Engineering, Chung Yuan Christian University, No. 200, Chung Pei Road, Chung Li, Taiwan

Introduction

Selenium (Se) is a naturally occurring trace element with a narrow exposure window between its beneficial and detrimental effects. Se pollution is a growing problem of global concern. The release of Se into the environment is associated with human activities, including agricultural practices, certain mining processes, the petrochemical industry, and high-tech industrial processes (Lemly 1997, 2004). Consequently, Se can rapidly attain levels that are toxic to the fish and wildlife in the aquatic and terrestrial ecosystems around the globe because of bioaccumulation in food chains and resulting dietary exposure. Se is an element essential to the health of humans, other animals, and some plants (Klasing 1998; Weeks et al. 2012). However, the

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behavior of Se is unusual in that Se also acts to cause adverse effects on reproductive success, which have been linked to declines in animal populations (Hamilton 2004; Lemly 1997, 2004).

The soil nematode *Caenorhabditis elegans* (*C. elegans*) is a ubiquitous soil organism that lives mainly in the liquid biofilm of soils. The genome of *C. elegans* shows a high level of conservation with the vertebrate genome, making *C. elegans* a good system for biological studies (Leacock and Reinke 2006; Schafer 2006). *Caenorhabditis elegans* is increasingly being utilized as a model organism in aquatic and soil toxicology due to its abundance in soil ecosystems and its properties of a short life cycle, ease of generating mass cultures, low cost of maintenance, and sensitivity to toxicants (Leung et al. 2008; Williams and Dusenbery 1990).

Due to the increasing concern regarding the environmental impacts of Se, the toxicological risks must be assessed with an ecological viewpoint. The interdisciplinary field of ecotoxicology attempts to assess the impacts of chemicals and other stressors on individuals, populations, and higher organizational levels by toxicological risk assessment with ecological perspectives (Billoir et al. 2009; Stark et al. 2004). Mathematical modeling can be a valuable tool for the prediction of population-level effects resulting from exposure to toxicants (Lopes et al. 2005). The toxic effect of a chemical on a population can be assessed by carrying out toxicity tests on the population itself. Alternatively, these consequences to a population can be inferred using mathematical models by incorporating information on survival, reproduction, and growth during the entire life cycle of an individual organism. The matrix population model (MPM) can be used to predict the future population dynamics by the maximum dominant eigenvalue, λ, i.e., population growth rate (Caswell 2001). Due to the biological relevance, the stage-classified matrix model has been applied widely in addressing the relationships among toxicants, in life-table response experiments, and in assessing population growth rates in ecological applications (Billoir et al. 2009; Chen and Liao 2004; Klok 2008; Liao et al. 2006; Lopes et al. 2005).

Given the vital but limited research on population outcomes in association with Se exposure and the potential ecological risks of the pervasiveness of Se in the environment, our objective was to use the nematode *C. elegans* as an ecological toxicity model to evaluate the toxicity impact of Se on the life cycle of the organisms. Herein, in the present study, the toxicity data associated with individual growth, survival rate, brood size, and hatching rate were used to determine the effects of Se(IV) on *C. elegans*. A stage-classified matrix model was applied to the toxicity data to assess the consequences of increased concentrations of Se(IV) on the population dynamics of *C. elegans* and to assess the effects of Se(IV) on the asymptotic population growth rate of *C. elegans*.

Materials and methods

Nematodes handling and reagents

Sodium selenite [Na_2SeO_3, Se(IV)] was purchased from Sigma-Aldrich (St. Louis, MO, USA). Wild-type *C. elegans* N2 (var. Bristol) and bacterial strains were supplied by the *Caenorhabditis* Genetics Center (CGC, University of Minnesota, MN, USA) funded by the NIH National Center for Research Resources. *C. elegans* was grown on nematode growth medium (NGM) plates using the *Escherichia coli* OP50 strain as a food source. A detailed *C. elegans* life cycle at 22 °C with discrete time points for each stage was previously described (Corsi 2006). Therefore, nematodes were cultured at 22 °C for further investigations based on the explicit time points of life cycle summarized by Corsi (2006). Worms were synchronized by hypochlorite treatment of gravid hermaphrodites (Sulston and Hodgkin 1998).

Survival and growth measurement

Survival and growth of the wild-type *C. elegans* were monitored from eggs to adults on NGM plates supplemented with various concentrations of Se (IV). The sodium selenite was dissolved in distilled water and then mixed into the liquid agar before pouring into the plates. The concentrations of Se(IV) tested were 0, 1, 2, and 3 mM (nominal concentrations). Synchronized eggs were incubated on each plate containing various concentrations of Se(IV) at 22 °C. The body length and survival of worms were scored at specific time points based on the normal life cycle of wild-type *C. elegans* at 22 °C according to the following scheme: 9 h (L1 larvae), 21 h (L2 larvae), 29 h (L3 larvae), 37 h (L4 larvae), 47 h (young adult), and 55 h (mature adult) after laying eggs (Corsi, 2006). The images of worms were captured using a Nomarski microscope (Leica, Wetzlar, Germany) and analyzed for the body length using ImageJ software (version1.40 g, National Institutes of Health, USA). The tests were independently performed 3–5 times. Approximately 20–30 worms were scored in each experiment.

Brood size and hatching rate measurement

Synchronized eggs were incubated on each plate containing various concentrations of Se(IV) at 22 °C until the L4 larvae stage. The L4 hermaphrodites were then placed individually onto the *E. coli* OP50 lawn on new NGM plates in the absence or presence of Se(IV) for continuous exposure. During the egg-laying period, individual nematodes were transferred daily to a new plate for 6 days until the egg-laying period was completed, and then, the eggs

were counted and summarized as total eggs. The number of the hatched L1 larvae counted was summarized and divided by the total number of eggs to determine the hatching rate. The tests were independently performed 3–5 times. Approximately 20–30 worms were scored in each experiment.

Modeling survival and growth toxicity data

Toxicity data on the life history of the survival and growth of worms were analyzed to link Se(IV) effects with model parameters. The stage-specific intrinsic survival rate in the control group [exposure to 0 mM Se(IV)] can be described by the Gompertz survival function (Yen et al. 2008) as:

$$S(t) = \exp\left[(B/G_s)(1 - e^{G_s t})\right] = 1 - M(t), \quad (1)$$

where $S(t)$ is the survival rate at time t, B is the initial mortality rate, and G_s quantifies the age-dependent acceleration and is referred to as the "Gompertz slope" (h^{-1}). For groups with the same Se(IV) treatment, a three-parameter Hill equation model (Hill 1910) was employed to express the stage-specific mortality rate:

$$M_i(t) = \left[\frac{M_{i,\max}}{1 + (LT_{50}/t)^n}\right], \quad (2)$$

where $M_i(t)$ is the mortality rate at time t, $M_{i,max}$ is the maximum mortality rate, LT_{50} is the time that causes 50 % mortality (h), and n is the Hill coefficient.

Individual growth was modeled with the von Bertalanffy growth function (Álvarez et al. 2005). The body length of individual *C. elegans* at time t, $L(t)$, was presented as:

$$L(t) = L_{\max}\left[1 - e^{-k(t-t_0)}\right], \quad (3)$$

where $L_{\max}$ is maximum body length (mm), k is the von Bertalanffy growth rate (h^{-1}), and t_0 is the theoretical age/time when the worm has a body length of zero (h).

Stage-structured matrix population model

To construct the stage-structured MPM of *C. elegans* exposed to Se(IV), six life stages based on the life cycle of *C. elegans* were selected. The six life stages were classified based on the time to reach the specific developmental stage under the normal life cycle of the wild-type *C. elegans* at 22 °C. Therefore, the six life stages (time) can be described as four larval stages: L1 (9 h) (including stage of egg), L2 (21 h), L3 (29 h), and L4 (37 h); young adult stage YA (47 h); and mature adult stage MA (55 h). The present matrix model applied a projection interval of 1 h ($T = 1$ h), starting with a certain number of individuals in each stage of population developed over time that can be represented as shown below (Caswell 2001):

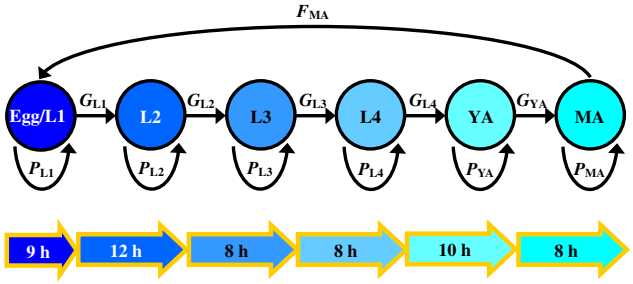

Fig. 1 A six-stage life cycle graph of *C. elegans*. The *circles* represent stages, and the *arrows* indicate transition probabilities: the probability of survival and remaining (P_i), the probability of survival and growth to next stage (G_i), and the fertility (F_i)

$$n(t+1) = \boldsymbol{A}n(t), \quad (4)$$

where n_i(t) denotes the abundance of *C. elegans* in stage i at time t, and matrix A is a population projection matrix. The stage-structured graph was shown in Fig. 1 with the transition probabilities, and matrix A was presented as follows:

$$\boldsymbol{A} = \begin{bmatrix} P_{L1} & 0 & 0 & 0 & 0 & F_{MA} \\ G_{L1} & P_{L2} & 0 & 0 & 0 & 0 \\ 0 & G_{L2} & P_{L3} & 0 & 0 & 0 \\ 0 & 0 & G_{L3} & P_{L4} & 0 & 0 \\ 0 & 0 & 0 & G_{L4} & P_{YA} & 0 \\ 0 & 0 & 0 & 0 & G_{YA} & P_{MA} \end{bmatrix}, \quad (5)$$

where P_i is the probability of survival and remaining in stage i, G_i is the probability of survival and growth from stage i to stage $i + 1$, and F_i is the fertility of stage i, the number of eggs produced per individual worm. The asymptotic population growth rate, λ, can be calculated as the maximum of the dominant eigenvalues of matrix A. If λ is greater than 1, the population increases over time, whereas if λ is smaller than 1, the population declines. When λ equals 1, zero net growth of the population results (Caswell 2001). The asymptotic population growth rate λ is related to the intrinsic rate of population increase, r, with $\lambda = \exp(r)$. P_i, G_i, and F_i are the transition probabilities in the population projection matrix A and are estimated from the toxicity data as (Caswell 2001):

$$P_i = \sigma_i(1 - \gamma_i), \quad (6-1)$$

$$G_i = \sigma_i\gamma_i, \quad (6-2)$$

$$F_i = f_e E, \quad (6-3)$$

where σ_i is the stage-specific survival probability in stage i (h^{-1}), γ_i is the growth probability (h^{-1}), f_e is the average number of eggs per individual per unit time (eggs h^{-1}), and E is the egg hatching rate (–).

The sensitivity of λ to changes in the transition probabilities provides an indication of which parameter has the

greatest effect on the population growth rate. Therefore, we can estimate

$$s_{ij} = \frac{\partial \lambda}{\partial a_{ij}}, \tag{7}$$

where *sij* is the sensitivity of a small change in the matrix element, a_{ij}, on λ (Caswell 2001).

Parameter estimation and model simulation

To estimate transition probabilities of the population projection matrix A, stage-specific survival and growth probabilities were estimated by using the toxicity data. The stage-specific survival probability σ_i in stage i was calculated by using the intrinsic survival rate and the Se(IV)-induced mortality rate as:

$$\sigma_i = \frac{S_i(t+T)}{S_i(t)T} \times [1 - M_i(t)], \tag{8}$$

where T is the projection interval (h), and the growth probability γ_i estimation was based on the body length as:

$$\gamma_i = \frac{L_i(t+T) - L_i(t)}{L_i(t)T}. \tag{9}$$

To analyze the uncertainty of model fitting and its effects on the estimation of asymptotic population growth rate, we implemented a Monte Carlo simulation that included input distributions for the parameters of the vital rate of *C. elegans* populations.

For estimates of F_i, the average number of eggs per individual per unit time (fe) was needed. The adopted laboratory data showed that wild-type *C. elegans* strains N2 had the maximum egg-laying rate of 141 ± 19 (mean $\pm$ SD) eggs d^{-1} with $N = 69$ individuals (Muschiol et al. 2009). The toxicity data of hatching rate for worms exposed to 0, 1, 2, and 3 mM Se(IV) were applied to E (egg hatching rate) of Eq. (6-3).

To simulate the stage-structured MPM of *C. elegans*, the present modeling used the initial number of *C. elegans* at six stages (n_{L1}, n_{L2}, n_{L3}, n_{L4}, n_{YA}, n_{MA}) that were arbitrarily assumed to be 200, 100, 100, 100, 100, and 100, respectively. Caswell (2001) indicated that the stable stage distributions and asymptotic population growth rate are not influenced by the initial condition.

Results

Toxic effects of Se(IV) on *C. elegans*

One recent study indicated that Se(IV) exhibits a dose- and time-dependent lethality on *C. elegans* (Morgan et al. 2010). Thus, to assess the population effects of Se(IV), several toxic effects were required to be examined for *C. elegans*. Growth, fertility, and survival were scored to represent the alteration of the population upon Se(IV) exposure. The concentration range of Se(IV) was adapted from Morgan et al. (2010) [0, 1, 2, and 3 mM Se(IV)].

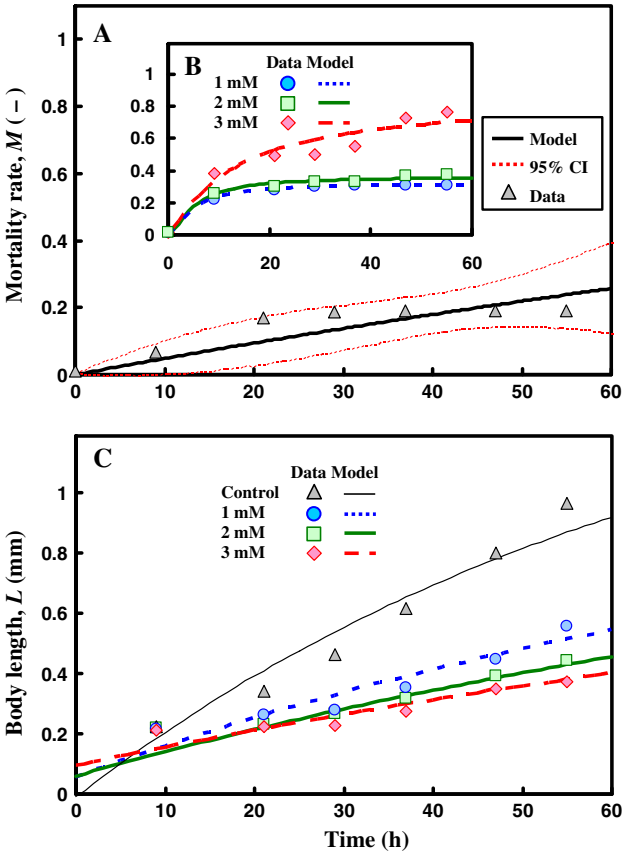

Fig. 2 Fitting performances of mortality and growth data for different Se(IV) exposure concentrations ranging from 0 to 3 mM. **a** The mortality rate fitted by the Gompertz survival function in the control group, **b** mortality rate fitted by the three-parameter Hill equation model for different Se(IV) treatments, and **c** time-dependent body length fitted by the von Bertalanffy growth function

Synchronized eggs were incubated on plates containing various concentrations of Se(IV) at 22 °C for the analysis of body length and survival. The results showed that Se(IV) resulted in dose- and time-dependent declines in growth and survival (Fig. 2).

To assess the reproductive effects of Se(IV), the brood size and hatching rate of worms were examined. The results showed that Se(IV) profoundly affected the production of *C. elegans*. Se(IV) significantly decreased both brood size and hatching rate of *C. elegans* (Suppl. Fig. 1). The values of brood size and hatching rate were 320 eggs and 99 % [0 mM Se(IV)], 54 eggs and 73 % [1 mM Se(IV)], 67 eggs and 74 % [2 mM Se(IV)], respectively, and 3 mM Se(IV) completely abolished the generation of progeny (Suppl. Fig. 1).

Table 1 Fitted model parameters for the Gompertz survival function, three-parameter Hill equation, and von Bertalanffy growth function

Trail	Fitting parameter			
	Gompertz survival function, S			
	B (−)	G_s (h^{-1})		r^2
Control	0.0049 ± 0.0026[a]	1.04×10^{-7} ± 0.024		0.64
Trail	Fitting parameter			
	Three-parameter Hill equation, M			
	M_{max} (−)	LT50 (h)	n	r^2
1 mM Se(IV)	0.32 ± 0.01	5.10 ± 0.65	1.46 ± 0.41	0.99
2 mM Se(IV)	0.36 ± 0.04	5.29 ± 1.81	1.50 ± 1.19	0.98
3 mM Se(IV)	0.85 ± 0.43	13.64 ± 13.96	1.10 ± 1.05	0.93
Trail	Fitting parameter			
	von Bertalanffy growth function, L			
	L_{max} (mm)	k (h^{-1})	t_0 (h)	r^2
Control	1.60 ± 1.59	0.014 ± 0.02	0.42 ± 7.65	0.94
1 mM Se(IV)	1.14 ± 2.23	0.010 ± 0.03	−5.00 ± 14.66	0.88
2 mM Se(IV)	1.08 ± 2.84	0.008 ± 0.03	−7.00 ± 17.93	0.81
3 mM Se(IV)	0.97 ± 3.10	0.007 ± 0.03	−13.99 ± 27.85	0.77

[a] Mean ± SE

Modeling survival and growth toxicity data

The mortality rate of *C. elegans* in the control group (no Se(IV) exposure) was fitted by the Gompertz survival function as presented in Fig. 2a and Table 1. Although the theoretical curve did not fit the data well ($r^2 = 0.64$), the toxicity data are still situated in the 95 % CI of the model (Fig. 2a, Table 1). The estimate of the Gompertz slope (G_s) was 1.04×10^{-7} h^{-1}. For groups of Se(IV) exposures, Fig. 2b shows the time-dependent mortality profiles of *C. elegans* described by a three-parameter Hill equation. In contrast to the non-exposed control group, exposed groups have a better fit in the mortality data ($r^2 = 0.93$–0.99) (Table 1). Moreover, the maximum mortality rates ($M_{i,max}$) increased as Se(IV) concentrations were elevated (Table 1).

The body length data were used to estimate the growth trajectories of the *C. elegans* life stages at different Se(IV) exposures by fitting the von Bertalanffy growth function (Fig. 2c, Table 1). As expected, both the maximum body length (L_{max}) and the von Bertalanffy growth rate (k) declined with the increase of the Se(IV) exposure concentrations (Table 1). The estimates of the maximum body length for *C. elegans* exposed to 0, 1, 2, and 3 mM Se(IV) were 1.60, 1.14, 1.08, and 0.97 mm, respectively (Table 1). According to the toxicity data and modeling results, growth inhibition is more sensitive for Se(IV) exposures than mortality, especially at a low concentration of Se(IV) (Fig. 2).

Vital rates and transition probabilities estimations

The stage-specific survival probability σ_i in stage i by Eq. (8) was estimated by linking the intrinsic survival rate with the time-dependent mortality rates for *C. elegans* exposed to the different Se(IV) concentrations. When *C. elegans* was not exposed to Se(IV), the survival probabilities were close to 0.99 h^{-1} for all stages. In contrast,

Table 2 Estimates of transition probabilities in population project matrix at six stages of *C. elegans* under different treatments of Se(IV)

	Control	1 mM Se(IV)	2 mM Se(IV)	3 mM Se(IV)
Stage	Transition probability			
P_i				
L1	0.88 (0.88–0.89)[a]	0.72(0.71–0.72)	0.70 (0.69–0.70)	0.64 (0.63–0.65)
L2	0.95 (0.95–0.95)	0.68 (0.68–0.69)	0.65 (0.65–0.65)	0.46 (0.45–0.46)
L3	0.96 (0.967–0.96)	0.68 (0.68–0.68)	0.64 (0.64–0.64)	0.39 (0.39–0.40)
L4	0.97 (0.97–0.97)	0.68 (0.67–0.68)	0.64 (0.63–0.64)	0.35 (0.35–0.35)
YA	0.98 (0.97–0.98)	0.67 (0.67–0.67)	0.63 (0.63–0.63)	0.31 (0.31–0.32)
MA	0.98 (0.98–0.98)	0.67 (0.67–0.67)	0.63 (0.63–0.63)	0.29 (0.29–0.30)
G_i				
L1	0.10 (0.10–0.11)	0.05 (0.04–0.05)	0.04 (0.04–0.04)	0.02 (0.01–0.03)
L2	0.04 (0.03–0.04)	0.02 (0.02–0.02)	0.02 (0.01–0.02)	0.01 (0.01–0.02)
L3	0.03 (0.02–0.03)	0.02 (0.02–0.02)	0.02 (0.01–0.02)	0.01 (0.01–0.01)
L4	0.02 (0.02–0.02)	0.01 (0.01–0.01)	0.01 (0.01–0.01)	0.01 (0.001–0.01)
YA	0.01 (0.01–0.01)	0.01 (0.01–0.01)	0.01 (0.01–0.01)	0.00 (0.00–0.00)
F_i				
MA	0.08 (0.06–0.11)	0.06 (0.03–0.09)	0.06 (0.04–0.10)	0 (0–0)

[a] Median (95 % CI)

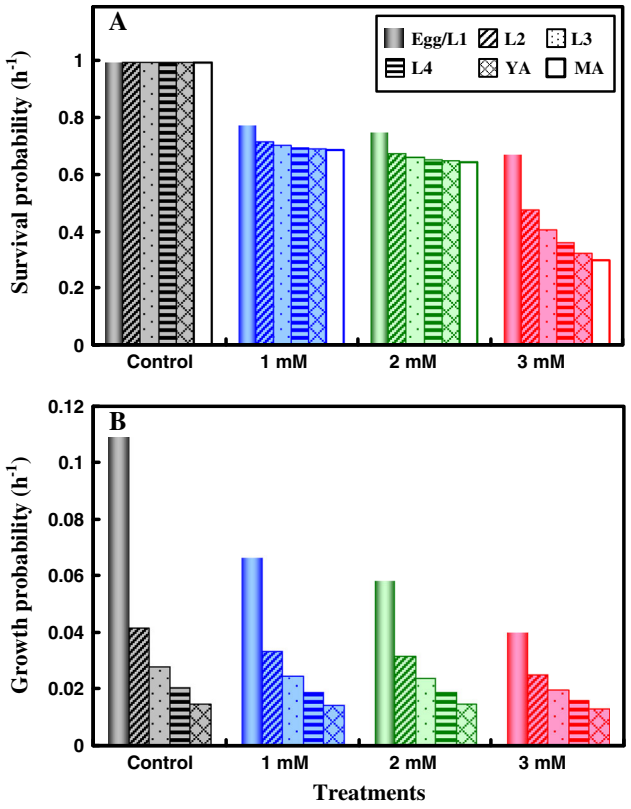

Fig. 3 Stage-specific survival (**a**) and growth (**b**) probabilities of *C. elegans* with exposure to different Se(IV) treatments

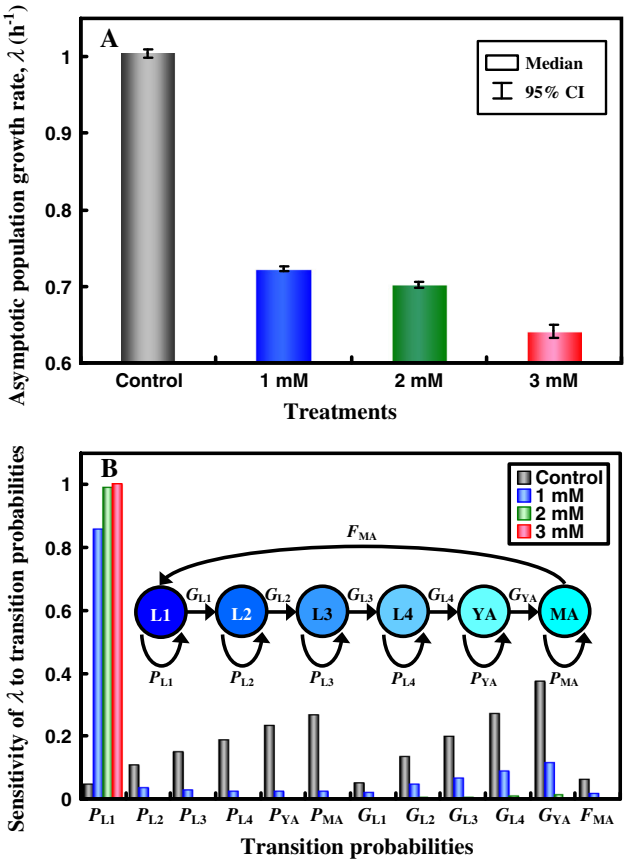

Fig. 4 **a** Asymptotic population growth rate (λ) of *C. elegans* with different Se(IV) treatments and **b** the sensitivity of λ to transition probabilities subject to different Se(IV) treatments

when the worms were exposed to Se(IV), significant decreases in survival occurred at all stages. The estimates of survival probability for worms exposed to 1, 2, and 3 mM Se(IV) from the life stage of L1 to MA were 0.77–0.69, 0.75–0.64, and 0.67–0.30 h^{-1}, respectively (Fig. 3a).

Figure 3b depicts the growth probability γ_i estimation based on the von Bertalanffy growth function. The growth probability of *C. elegans* for the L1 stage has the largest decline from 0.11 h^{-1} (for the control) to 0.04 h^{-1} (for exposure to 3 mM). In contrast, there was little effect on the growth of *C. elegans* at the young adult stage with a range from 0.013 h^{-1} to 0.015 h^{-1} (Fig. 3b).

Estimates of the probability of survival and remaining (P_i) and the probability of survival and growth to next stage (G_i) can therefore be obtained using Eqs. (6-1) and (6-2). The fertility (F_i) was estimated by incorporating hatching rates obtained with toxicity data into the fecundity that was adopted from Muschiol et al. (2009). For the control group, P_i increased from stages L1 to MA, whereas P_i showed a decrease for the Se(IV) treatment groups (Table 2). G_i declined with increasing developmental stage and Se(IV) concentrations (Table 2). Due to the toxicity, the hatching rate was 0 % for *C. elegans* exposed to 3 mM Se(IV), and fertility was also equal to zero (Table 2).

Asymptotic population growth rate

The toxicity tests for survival, growth, and reproduction for *C. elegans* were further introduced into the six-stage MPM to estimate the asymptotic population growth rate (λ) for different Se(IV) treatments. Figure 4a demonstrated that the asymptotic population growth rate decreased significantly with the increased Se(IV) levels. When *C. elegans* was not exposed to Se(IV), the value of $\lambda = 1.00$ (95 % CI: 0.99–1.00) h^{-1} was used as the baseline to compare with the value of λ for each Se(IV) treatment. The estimated values of λ were 0.72 (95 % CI: 0.72–0.72) h^{-1}, 0.70 (95 % CI: 0.69–0.70) h^{-1}, and 0.64 (95 % CI: 0.63 – 0.65) h^{-1} for 1, 2, and 3 mM Se(IV), respectively, indicating that λs are significantly lower than the values in the control group (Fig. 4a). A potential risk of decreased population for *C. elegans* exposed to a Se(IV)-contaminated environment therefore exists.

The sensitivities of population growth rate to the transition probabilities (P_i, G_i, and F_i,) exhibit an obvious difference between the control and the Se(IV) treatment groups (Fig. 4b). The sensitivities of λ to P_i and G_i

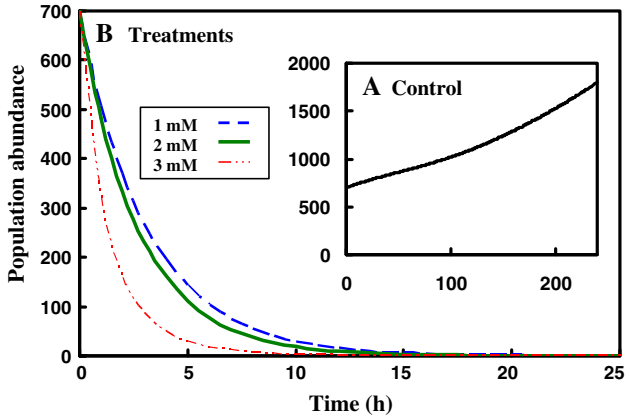

Fig. 5 Temporal changes in overall population abundance of *C. elegans* in **a** control group and **b** Se(IV) treatment groups

increased with developmental stage in the control group, and the most significant stage was the probability of survival and growth at the young adult stage (G_{YA}) (Fig. 4b). In contrast, P_{L1} was the most sensitive parameter for all Se(IV) treatment groups (Fig. 4b).

Population abundance

Figure 5 illustrates the time influence on the population abundance of *C. elegans* exposed to the different concentrations of Se(IV). In the control group, the abundance of *C. elegans* followed an increasing curve after 10 days with nearly 1,800 individuals (Fig. 5a). The population of *C. elegans* for all Se(IV) exposure groups decreased sharply from 700 to 0 individuals within 1 day. According to the modeling simulation, the population will become extinct at 21, 19, and 13 h while exposed to 1, 2, and 3 mM Se(IV) (Fig. 5b). However, compared with the experimental data, the risk of reduced population growth seems to be overestimated by the modeling simulation.

Discussion

Mathematical modeling approaches can be a valuable tool to clarify the complex interactions between chemical compounds and individuals, between the physiological endpoint and the life table traits and between individuals and populations in which the ecotoxicological outcome can be supposed to be predictive and not only descriptive. Although MPMs are not novel in ecotoxicology, a significant gap remains between the theoretical structure and the exploration of the models and their application to specific ecotoxicological questions and management problems. The limited application of these models to realistic problems has many explanations, including: (i) confusion over the relationship among models, particularly between the life tables and the projection matrix, and (ii) difficulties in parameterizing models with reliable estimates of vital rates in the population.

The simultaneous fits of the model are shown in Fig. 2 and Table 1 for the required parameters. For the analysis of survival, some misfit of the survival data occurred in the control group. The Gompertz slope (G_s) obtained at 2.50×10^{-6} d^{-1} in our estimation is considerably less than in previous studies by 4–5 orders of magnitude (Brooks et al. 1994; Lenaerts et al. 2007; Vanfleteren et al. 1998; Wu et al. 2008). The differences between our estimate and the reference data may be related to the differences in experimental lengths of time (Brooks et al. 1994; Vanfleteren et al. 1998), possibly due to the experimental period of survival data at 55 h in the present analysis, whereas for previous studies, the period was 20–50 days.

The widely used von Bertalanffy growth model is especially fit for short life-span organisms (Cartaxana 2003). In the control group, the von Bertalanffy growth rate (k) of *C. elegans* is equal to 0.014 ± 0.02 h^{-1}, which is close to the value of 0.016 h^{-1} (Wren et al. 2011). Our value is 30 % lower than the rate of 0.020 h^{-1} estimated by Álvarez et al. (2005) and considerably lower than the rate of 0.039 h^{-1} calculated by Jager et al. (2005). *C. elegans* has an S-shaped growth curve—an exponential phase of larval growth and a gradual approach to a plateau in late adulthood (Knight et al., 2002). For the same reason, the 55-hour experimental period may not completely describe the manner of growth for the entire *C. elegans* lifespan.

In this work, we demonstrated that Se(IV) has a profound impact on the population dynamics of *C. elegans*. Under Se(IV) exposure, we observed increased mortalities, shortened body lengths, and damaged fertilities of *C. elegans* (Fig 2 and Suppl. Fig. 1). Boehler et al. (2013) suggested that maturation delay was the likely source of the body length decrease by Se(IV). We observed that Se(IV) significantly delayed the developmental stages of nematodes after 9 h by comparing with the non-exposed control nematodes (Suppl. Table 1). Moreover, Boehler et al. (2013) reported that developmental arrest could be observed under 1 mM or 2 mM Se(IV) treatment in axenic media. Estevez et al. (2012) showed a reduction in the egg laying rate after Se(IV) exposure, but did not look at overall brood size or hatching rates. This might be due to the short exposure time to Se(IV) (1 to 6 h exposure followed by 24 h recovery). In the present study, we incubated nematodes with Se(IV) in the presence of *E. coli* OP50 continuously from eggs and found that several worms could still reach the L4 stage or even adult stage to generate the progeny. The discrepancy in Se(IV) toxicity to *C. elegans* might be due to bacterial metabolism from

E. coli OP50. It has been reported that the bacteria can metabolize the drug compound before it reaches the target site of nematodes and alters the observable phenotypes (Kaletta and Hengartner 2006; Saiki et al. 2008). In addition, Morgan et al. (2010) reported that adult nematodes exhibited reductions in motility regardless of whether they were grown in axenic medium or in the presence of *E. coli* OP50. Therefore, *E. coli* OP50 on NGM plates likely metabolized Se(IV) to the less toxic Se(0) (Nuttall 2006; Li et al. 2011). In the environment, bacteria are likely to be present in soils, we therefore included *E. coli* OP50 during the Se(IV) exposure periods in order to reflect the toxicological risk of Se on *C. elegans* in the ecosystems.

The asymptotic population growth rate (λ) was estimated as ranging from 1.00 to 0.64 h^{-1} for increasing Se(IV) concentrations. The population became extinct when λ fell below a value of 1. Our λ estimates cannot be compared with the λ estimates from Chen et al. (2006) (3.85 d^{-1}), possibly due to the difference in projection intervals (1 h in our analysis, but 1 day in Chen et al. (2006). Consistent with Chen et al. (2006) and Lopes et al. (2005), we found that the asymptotic population growth rate was sensitive to change in larva survivorship for Se(IV) treatment. Because larvae become more tolerant with increasing age, this result implied that survivorship of L1 larvae was a significant factor. Part of the reason for the magnitude of this difference for the population growth rate is exhibited by *C. elegans* under different laboratory culture conditions. Care is needed in making evolutionary inferences from such ecologically unrealistic conditions (e.g., food source and quantity, predator-free environment, space limitation, temperature, etc.) (Chen et al. 2006).

In principle, this work showed how physiological responses to Se(IV) at the individual level can be related strictly to predicted population dynamics responses. The prediction can be achieved in a straightforward manner by survival rate, growth, brood size, and hatching rate. The MPM does not take into account population effects that occur through interactions between individuals: migration, competition, predation, food availability, or other factors that affect population growth. These interactions are likely to be of highest importance in populations. Through this work, an important limitation is that we assumed that the exposure periods of all toxicity data were sufficient to illustrate the effects on the whole life history of the *C. elegans* population. The prediction of the field effects by directly utilizing "population impacts" that were estimated from a population model remains questionable. The population project matrix does not consider mixed stress under field conditions or density-dependent effects. Lin and Meng (2009) nevertheless noted some implications associated with using the MPM for management of a contaminant: namely, a population-based decision-making applying to ecological/environmental risk assessment, a cost-effective approach, ease of risk comparison, and reasonable extrapolation by integrating acute and chronic toxicity data. All the endpoints are integrated into an ecologically relevant parameter, a crucial step forward from the current practice in assessment of ecotoxicological effects (Jager et al. 2004).

In conclusion, the present simulation represents how a mechanistic view based on the effects of Se(IV) on the soil nematode *C. elegans* can promote life cycle toxicity assessment. The MPM could help in understanding the toxic effects of Se(IV) at the *C. elegans* population level on the basis of toxicity data analyses at the individual level and the cooperation of individual effect models relating the life table traits to the exposure to Se(IV) concentrations. An important implication of the analysis is that mathematical models can be used to give a population stage structure and to give clarity to the analysis of the key population-level endpoint (the asymptotic population growth rate) of population dynamics, as well as to evaluate the influence of the response of other species to environmental Se(IV). These models sequentially provide candidate environmental criteria for the evaluation of the population impact of Se.

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