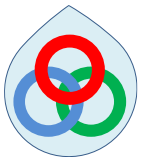

Manual and Guidance of A-TERAM

Content

Preface － Outline of “A-TERAM” and the Manual

This document describes the operation method and the general background of Aquatic Tri-trophic Ecological Risk Assessment Model (A-TERAM) and its software A-TERAM version 1 developed to evaluate the impact of chemical substances on aquatic ecosystems.

In the field of ecological risk assessment, which assesses the impact of chemical substances on ecosystems, how to calculate the standard that can reasonably achieve ecosystem conservation from toxicity values that indicate decreased survival and reduced fertility measured at the individual level has been taken up as a major issue (Ferson et al. 1996; Pastorok et al. 2002; European Commission 2003). At assessing the impact on ecosystems, what is set as the final protection goal and the extent to which ecological complexities such as interspecies interactions between organisms and so on should be reflected may be common issues (Bartell et al. 1992; Suter and Barnthouse 1993; Hommen et al. 2010; Menzie et al. 2008). Furthermore, in addition to such ecological relevance, in order to support the management and regulation of chemical substances, versatility applicable to many chemical substances, consistency of risk assessments across chemical substances and transparency that clearly shows the assumptions and calculation process of risk assessment are required for actual implementation (Galic et al. 2010).

A-TERAM has been developed with the aim of addressing these issues. It is constituted by three trophic levels including primary producers (algae), primary consumers (*Daphnia*), and secondary consumers (fish). The basic structure of such an ecological model of A-TERAM corresponds to the basic ecotoxicity testing using the test species belonging to these trophic levels, which have been conducted in many countries as standard ecotoxicity tests for assessing the ecological effects of chemicals. On the other hand, limnology and aquatic biology have pointed out that the food chain through large Cladoceran (*Daphnia*) is an important basic structure for maintaining the function of aquatic ecosystems such as lakes and marshes (Andersen 1997; Horne and Goldman 1999).

In order to reflect the ecological complexity in the risk assessment, A-TERAM mainly incorporates two ecological factors. One is the introduction of interspecies interactions (Begon et al. 2013) based on the relationship between fish and *Daphnia* and between *Daphnia* and algae. This is one of the most important differences from many conventional ecological risk assessment methods. In the actual natural ecosystem, algae of the primary producer, which is the only trophic level that can produce organic matter by photosynthesis, are consumed by *Daphnia* as the primary consumer, and *Daphnia* are eaten by fish as the secondary consumer. Predation establishes the flow of organic matters, nutrients and energy (Andersen 1997; Cebrian 2004; Dickman et al. 2008). These ecosystem processes are necessary for the function and health of ecosystems to be maintained.

Another ecological factor is difference in life history characteristics and seasonal (phenological) schedules due to different trophic levels and species identities. The species groups that make up the three trophic levels not only have different functions in ecosystems, but also belong to very different taxonomic groups and have diverse life history characteristics (survival rate, growth rate,

age of first reproduction, reproduction period, fecundity, life span, etc.). For example, *Pseudokirchneriella subcapitata*, the typical unicellular green alga (phytoplankton) designated as a test species under the Chemical Substances Control Law of Japan, is capable of multiplying by 10 folds in a few days by cell division, but on the other hand, many freshwater fish species like medaka (*Oryzias latipes*), which breed only once in a year, in spring, and grow over several months, increase the population number only several times at most over a year in the field. In addition, in aquatic ecosystems such as lakes and marshes, due to differences in the life history characteristics of composite species, seasonal changes in species abundances and constitution are commonly observed such as algae bloom in the early spring, lake water clear phase in the spring owing to the proliferation of Daphnia, and fish breeding and growth from spring to summer (Brönmark and Hansson 1998; Horn Goldman 1999).

The most common large zooplankton like *Daphnia* spp in static aquatic environment, which are thought to be also important in supporting aquatic ecosystems, appear mostly in the spring when water temperature is low. It is known that they spend summer usually as resting eggs in dormancy. Considering these ecological factors, the ecological significance and effects can vary from species to species, even with the same toxic endpoints such as mortality and reproductive inhibition, and the environmental concentrations of chemicals. It is inferred that the effect evaluation of chemicals may differ depending on whether the seasonal changes in biota are taken into account, because species composition in community changes seasonally.

When assessing the impact of chemicals on ecosystems, the major issue is what to set as the final protection goal. The final protection goal must be ecologically sound and relevant with enough consensus among policy makers and stakeholders. The efficiency in regulation of ecological risk assessment is limited unless the methods and results of risk assessment are clearly and quantitatively presented.

The final protection goal of A-TERAM is to preserve species diversity and maintain ecosystem functioning. As ecological risk assessment criteria for achieving these protection goals, we focus on the population growth rate of fish, the highest species in the three trophic levels, and calculate the proportional reduction of the population growth rate due to chemical substances as an ecological risk quotient. It is well known that the probability of extinction, the ultimate hazard for a population, depends greatly on the rate of population growth (Lande 1998; Lande et al 2003). Decreases in population growth due to pollutant chemicals can be translated into population or species extinction risk (Tanaka and Nakanishi 2000; Nakamaru et al. 2002; Tanaka 2003).

As a criterion for achieving the protection goal that is conservation of species diversity, we focus on the population growth rate of fish and consider the viability of algae and *Daphnia* populations as not a direct criterion. It is inferred not practical to estimate the risk of species or population extinction in these species, because most zooplankton and phytoplankton (green algae, blue-green algae, crustacean, etc.) that inhabit lakes, marshes, ponds and drainage channels in temperate regions are cosmopolitan species which are distributed over a wide range on the earth. Very few species are designated as endangered. On the other hand, many freshwater fish, including medaka *O.*

latipes, have been designated as endangered species, and it is considered that there is some reality in estimating the extinction risk for such species.

In natural ecosystems, algae are responsible for primary production, while *Daphnia* support ecosystems in terms of trophic flow. In other words, algae produce organic matter from inorganic matter by photosynthesis and feed themselves to primary consumers to support the conversion of organic matter and productivity at higher trophic levels in ecosystems. *Daphnia*, as a major herbivore of aquatic ecosystems, ingests algae efficiently and transfers the organic matter produced by the algae to species at higher trophic levels (Andersen 1997; Hanazato 1998). It is thought that the maintenance of such nutrient or energy flows through food chains is essential for preservation of ecosystem health and functioning.

A-TERAM hypothesizes that the toxic effects of chemicals on the ecosystem functions of algae and *Daphnia* are also represented by a decrease in the population growth rate of the fish, which occupies at the highest level in the model. This is because of the role played by the algae and the *Daphnia* in ecosystems. The primary productivity by the algae does not transfer to higher levels nor base the aquatic ecosystem until the algae are ingested by *Daphnia* and the *Daphnia* is eaten by the fish. Similarly, the role of Daphnia in material flow in ecosystems can be assessed as its effect on the population growth rate of the planktivorous fish (Kemp et al. 2001; Tanaka and Mano 2012).

In other words, in A-TERAM, the effect of chemicals that attenuates *Daphnia* populations is evaluated as a decrease in the population growth rate of the fish. Similarly, the effect of chemicals that inhibits algae population growth is assessed by attenuating the *Daphnia* which graze algae. Finally, by the fish reproduction diminished by lack of food. Therefore, A-TERAM unifies the ecological risks associated with the two protection goals of preserving biodiversity and preserving ecosystem function as a decrease in the population growth rate of fish, the top species in the tri-trophic model.

In the risk assessment and management frameworks for chemical substances, the utilities of the mathematical model like A-TERAM could provide are constrained by the toxicity and other chemical information the model requires and the attributes of the outputs by the model. The traditional risk assessment method, the hazard quotient method, is based on the ratio of PEC (predicted environmental concentration) to PNEC (predicted no effect concentration), which is the smallest NOEC (no observed effect concentration) among the test species divided by the uncertainty factor UF. This method is relevant for the purpose of the screening-level assessment, because it regulates environmental concentrations of chemicals towards lower than the marginal level that is considered to induce no hazardous chronic effect to all test species and most species at the three trophic levels in the wild. On the other hand, A-TERAM is suitable for risk assessment in situations where there are potential risks for environmental concentrations of chemicals to exceed PNEC. For example, within the framework of the Chemical Substances Control Law, A -TERAM may be applied to the chemicals with relatively high-risk priorities at the screening level and the risk assessments after the first-tier (stages II and III of the primary risk assessment).

The traditional framework calls for a quantitative or semi-quantitative assessment of the

ecological risk of chemicals (risk ranking) at these stages of the risk assessment. However, it must be based on the poorly quantifiable PNEC. On the other hand, if an ecological model is available, it is necessary to ensure consistency of the risk assessment between the screening level and higher stages of the risk assessment based on the ecological model. We have to consider the limitation that many chemicals hardly collect more ecotoxicity data than the minimum data that the first-tier screening requires. A-TERAM has been developed as a system that satisfies these conflicting demands and realizes quantification of ecological risk assessment by applying the basic principles of ecology to the framework of public ecotoxicity testing.

For consistency in the risk assessment method for various chemicals which have different extents of ecotoxicity data it is necessary to be able to perform risk assessment with a minimum set of ecotoxicity data and at the same time to be able to use extensive ecotoxicity data if available for risk assessment. Basically, the same theory is applied to a variety of chemicals, including chemicals in which the minimum screening-level ecotoxicity information is available and chemicals in which a plenty of data are available thereby detailed ecological risk assessment is feasible. Without a uniform framework for assessing risk, it is not possible to assure the consistency of risk assessment methods. For example, even if complex ecological models are very elaborately designed to mimic natural ecosystems and are effective in precise estimation of risks, it would not be possible to confirm the consistency with risk assessment methods for many chemicals, if the required ecotoxicity information is so enormous that the application is limited to a few intensively studied chemicals.

As an effort to address the above issue, A-TERAM can work with the basic ecotoxicity data at the three trophic levels (at least one species from each level) and exposure information, while A-TERAM can use much of the ecotoxicity data consistent with the OECD test guidelines for protecting freshwater ecosystems. The minimum required ecotoxicity information (the essential data) for A-TERAM is the fish acute toxicity (half-lethal concentration, LC_{50}), the *Daphnia* acute immobilization (50% effect concentration, EC_{50}), and the algal growth inhibition (no effect concentration NOEC or 50% effect concentration, EC_{50}).

In addition, A-TERAM allows you to enter toxicity values for the fish growth inhibition (NOEC), the fish reproductive inhibition (NOEC), and the *Daphnia* reproductive inhibition (NOEC). The fish growth inhibition is a chronic toxicity that is estimated from a test method called the early life stage test. A-TERAM expresses the ecological effects of fish growth inhibition as a decrease in the number of eggs laid by fish of reproductive age. The *Daphnia* reproductive inhibition examines decreases in the total number of offspring delivered by each mature female when *Daphnia* are raised under chemical exposure conditions for a long period (usually 21 days) from the beginning of life history.

A-TERAM does not exclusively use ecotoxicity data according to a specific test method, but the ecotoxicity information which includes ecotoxicity tests results according to the following OECD Test Guidelines (TG) can be input.

Fish acute lethal effect：TG203

Fish growth inhibition：TG210
Fish reproductive inhibition：TG229
Daphnia acute immobility：TG202
Daphnia reproductive inhibition：TG211
Algae growth inhibition：TG201

These ecotoxicological information almost completely covers common practices of ecotoxicological tests aimed at preserving freshwater ecosystems. In A-TERAM, these ecotoxicities are attributed to individual-level life history characteristics (fish survival, fish growth rate, fish fecundity, *Daphnia* survival and reproduction, and algal growth), which exposure to certain chemicals (with certain concentrations in the environment) affects. The toxicity information is used to quantify the extent to which adverse reactions can be caused by a particular exposure level.

If A-TERAM lacks the essential ecotoxicological information required to operate, namely the acute fish toxicity, the *Daphnia* acute immobility, and the algal growth inhibition, A-TERAM processes acute-chronic extrapolation. Automatically it estimates the missing ecotoxicity in the three essential data and calculates the ecological effects in the same way as when all ecotoxicities are available. Specifically, the NOEC of fish growth inhibition and reproductive inhibition are extrapolated from the LC50 of fish acute toxicity, and the NOEC of *Daphnia* reproductive inhibition from the EC50 of *Daphnia* acute immobility. When chronic toxicity values are available, A-TERAM gives priority to the input chronic data over the indirect estimate extrapolated from acute data.

The most common method of acute chronic extrapolation is to divide the acute toxicity value by a predetermined index (ACR: acute-chronic ratio). A-TERAM developed an extrapolation model (a regression equation) based on the database of a project, “Results of Ecotoxicity Tests of Chemicals”, which was conducted by Ministry of the Environment (http://www.env.go.jp/chemi/sesaku/02e.pdf). This database is referred to as “Ecotox-MoE” in this document. Ecotox-MoE is a collection of test results for a wide range of chemicals, commissioned by Ministry of the Environment by a GLP testing institutes. It is considered to be one of the best sources of ecotoxicity data for our purpose in that they were estimated under uniform test conditions using standard test species.

Besides the ecological factors that are explained before, A-TERAM takes into account the long-term adverse effect of pollutants that could be facilitated by bioaccumulation of the pollutants. For this purpose, it requires information concerning the bioconcentration of chemicals as necessary input data.

If the accumulation of a chemical in an organism is high and it takes time for the concentration of the chemical in the organism to reach the maximum concentration (equilibrium concentration), the toxicity estimated in the short-term acute test may be underestimated for toxicity by long-term continuous exposures. It has been pointed out that the toxicity value (LC_{50}, etc.) of a highly bioaccumulative chemical is time-dependent (the longer the test period, the smaller the toxicity value and the stronger the toxicity) (Sprague 1969; Suter 1993).

A-TERAM incorporates the time dependency of ecotoxicity due to the accumulation of chemicals only for fish. The effect of bioaccumulation on algae and *Daphnia* was not considered. It could be inferred that the longevity of these species in the field is unlikely to be noticeably longer than that of common ecotoxicity test periods and then the effect of bioaccumulation of chemicals on the toxic response by these species is considered to be limited. Another reason for neglecting bioaccumulation in algae and *Daphnia* is little information available on bioaccumulation in these species.

In A-TERAM, the emphasis was put on balancing versatility, consistency and transparency, with the aim of reflecting ecological relevance in the risk assessment. Therefore, some of the factors that could be important in assessing the ecological risks of chemicals were omitted on purpose. I would like to mention how these factors should restrict the application of A-TERAM.

One of the omitted elements is interspecies extrapolation of ecotoxicity values. According to the official ecological impact assessment methods under the Chemical Substances Control Law of Japan, in case hazard information on one of the three trophic levels is not available the toxicity value of the missing trophic level is extrapolated from the toxicity values of other trophic levels by interspecies extrapolation. However, A-TERAM does not include a module for interspecies extrapolation, so it is necessary to input at least one of the acute or chronic data for all three trophic levels. In order to refine the setting of uncertainty factors for interspecies extrapolation, it is necessary to practice comparative toxicological surveys to establish the best statistical estimate of uncertainty factor by interspecies extrapolation, which is beyond the task of A-TERAM. A-TERAM focuses on implementing ecological risks that take into account ecological factors and leaves the uncertainty issues arising from interspecies extrapolation.

The effect of biomagnification of chemicals through the ecological food web are not incorporated into the model although the effect of bioconcentration of chemicals on chronic toxicities at the level of fish individuals is embedded into the model. In other words, the model does not include the process in which persistent and highly bioaccumulative chemicals are gradually concentrated in the body of organisms at higher trophic levels through the predator-prey relationship between species at different trophic levels. The hazardous chemicals with extremely high bioaccumulation and little biodegradability such as persistent organic pollutants (POPs) are not the target chemicals of A-TERAM for conducting ecological risk assessment.

A-TERAM uses an ecological model which assumes that each of the three trophic levels is occupied by one species. However, in a real ecosystem, multiple species coexist at each trophic level. In addition, interspecific relations are caused not only by vertical relations (predator-prey relationships) assumed in A-TERAM but also by horizontal relations (interspecific competition for resources). In addition, there are a variety of interspecific relationships such as intraguild predation caused by omnivory of the top species and apparent competition which is caused by shared predators. These ecological complexities are not included in A-TERAM.

Regarding ecotoxicity data, toxicity values may be collected for many species for the same chemical substance, and the species sensitivity distribution (SSD) may be available. In particular,

as the stage of risk assessment is higher, ecotoxicological information tends to be collected for a wider range of species. How to incorporate it into the future regulatory ecological risk assessment is a future issue.

The last ecological factor that A-TERAM does not address is the spatial distribution of organisms and chemicals, or the spatial structure of populations and communities. Several studies have suggested that the spatial structure of populations and communities can influence the ecological risk of chemicals. However, the environment assumed by A-TERAM is a small, closed static water area such as ponds and agricultural drains, and the target species (algae, daphnids, and small freshwater fish) do not have high migratory ability across large spatial scales. For this reason, it is not likely that the populations of these species have a dynamic spatial structure such as meta-populations and meta-communities. If we address the issue of the spatial distribution of ecological risks in relation to the local environmental concentrations and exposure assessments of chemicals, we may assume independent aquatic ecosystems for different exposure sites and may have to add up the calculation results of A-TERAM from each site to derive a large-scale risk estimation.

Finally, A-TERAM is not proposed as an established tool for ecological risk assessment, rather integrates knowledge from different fields including environmental chemistry, ecotoxicology and ecology. I would like to mention that A-TERAM has been proposed as a platform to promote exchanges of expertise from different fields. In fact, some assumptions and calculations that affect the results of the risk assessment have not been fully considered by experts in each field. We believe that the presented model will be further improved through interdisciplinary research and arguments, thereby providing a template for more complete methods.

A-TERAM was created with the cooperation of many experts. Dr. Hiroaki Shiraishi, Dr. Yasunobu Aoki and Dr. Noriyuki Suzuki of Center of Health and Environmental Risk Research, National Institute for Environmental Studies supported the development of A-TERAM through the planning and financial support of research projects as department heads. Dr. Kenichiro Sakurai of the center provided expert knowledge on bioaccumulation of chemical substances, and Dr. Shigeto Oda assisted in collecting and organizing ecotoxicity information. Mr. Kazuo Hasunuma of Center for Health and Environmental Risk Research and Mr. Takehiko Fukushima and Mr. Ryosuke Takahashi of the Ministry of the Environment, Environmental Policy Bureau, Environment and Health Department (both on January 2016) permitted us to use the original data of Ecotox-MoE for our convenience. Also, Takeo Nakamura, a software engineer, was essential in developing A-TERAM software. Without the support of these people, A-TERAM would not have been able to finalize the form that could be used as a tool to support ecological risk assessment.

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Part I　Manual

1 - 1　Installation of the software

A-TERAM ver.1 can be installed on MS-Windows 7 or later. The system requirement is at least 180 MB of extra disk space.

To install, double-click the installer A-TERAM Setup and follow the on-screen instructions.

1 - 2　Preparation of input data

The essential ecotoxicological data for calculating ecological risk are the fish acute toxicity LC_{50} and the test (exposure) duration, the *Daphnia* acute toxicity (immobility) EC_{50} and the test (exposure) duration, and the algal growth inhibition NOEC or EC_{50}. Furthermore, the chemical concentration in the environment, the bioconcentration factor BCF, and if BCF is equal to or larger than 100, the elimination constant k_e or the octanol-water partitioning constant K_{ow} are required.

In addition to the above three kinds of ecotoxicity data, you can also enter NOECs for the fish growth inhibition, the fish reproduction, and the *Daphnia* reproductive inhibition.

A-TERAM itself does not perform interspecies extrapolation. Acute toxicity values (NOEC values are also possible for the algae) are necessary at all three trophic levels. If there is a trophic level for which toxicity information is missing, enter an indirect value that is estimated by the interspecific extrapolation, the QSAR (the quantitative structure-activity relationship) method or other methodscvf.

1 -3　Main screen

When you start A-TERAM, you see the main screen shown in Figure 1-1 displayed first. On the main screen, you can manage the data of chemical substances (new input, save and load), operate simulations, and set detailed model parameters.

The box that occupies the upper half of the main screen lists the substance files that are being referenced. You can enter a new substance file or open an existing file. To perform either operation,

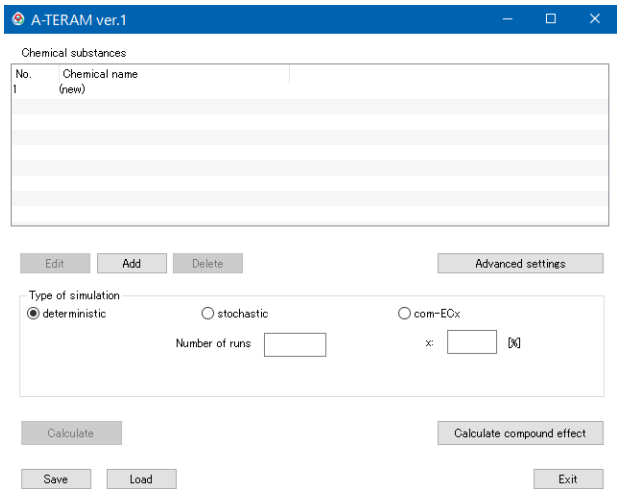


Figure 1-1　Main screen

place the cursor on a chemical substance (No. 1 (new) in Fig. 1-1) in the list and select it by left clicking it to highlight blue.

Next, click Edit to enter new data, or click Read to read from a saved file.

1 - 4 Input ecotoxicity information and bioconcentration data

Ecotoxicity data, bioconcentration data, and environmental concentration settings for chemical substances are stored in the chemical substance file for each chemical substance.

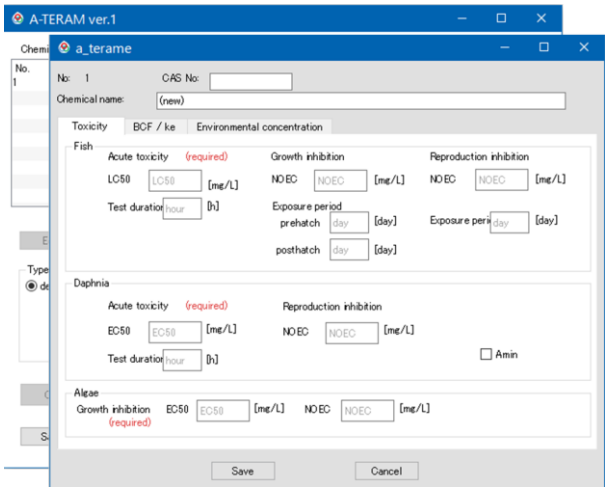


Figure 1-2 “Toxicity value” sheet in the chemical substance file

This section describes how to enter new information on ecotoxicity and bioaccumulation of chemical substances. Highlight “(New)” from the substance list on the main screen and click Edit. A new chemical substance file opens as shown in Figure 1-2. The chemical substance file consists of three sheets: "toxicity value", "BCF / ke", and "environmental concentration". Figure 1-2 shows a toxicity value sheet is opened for a chemical substance file.

First, enter the chemical substance name, or CAS number (optional). Next, enter all toxicity values in the unit of mg/L. Fish acute toxicity, *Daphnia* acute immobility, and algal growth inhibition (at least one of EC_{50} and NOEC) are essential data. For the fish acute toxicity and the *Daphnia* acute toxicity, a test duration (hours) is also required. If you are unsure about the test duration, we recommend you enter the most common test durations, say 96 [hr] or 48 [hr].

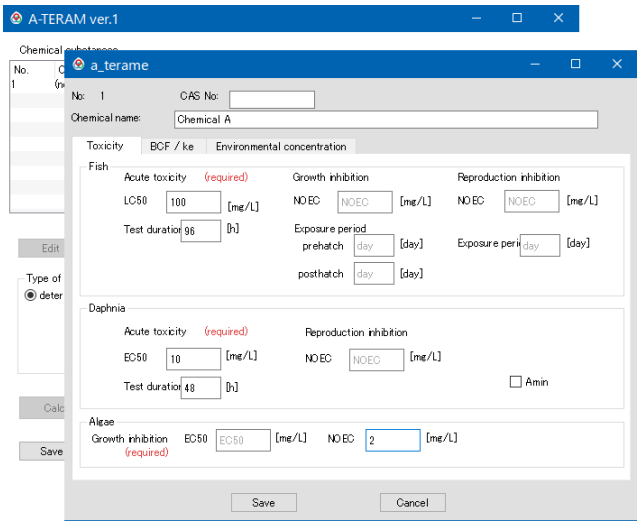


Figure 1-3 A screen illustrating input of toxicity data of Substance A in the toxicity value sheet

If you have data on fish growth inhibition or reproductive inhibition, enter the exposure duration in days. For the growth inhibition test results, enter the exposure period separately before hatching and after hatching. If there is no exposure before hatching, you can enter 0 for the exposure concentration before hatching.

If there is no data on *Daphnia* reproductive inhibition, check the amines box if the target chemical is an amine. A-TERAM performs the acute chronic extrapolation of *Daphnia* after distinguishing amines from non-amines.

If there is no data other than the required ecotoxicological data, leave the input box blank. If there are some blanks, A-TERAM performs the acute-chronic extrapolation. If a value greater than 0 is entered, it will be considered as a measured chronic toxicity value and will be used as an ecotoxicity datum prior to extrapolation.

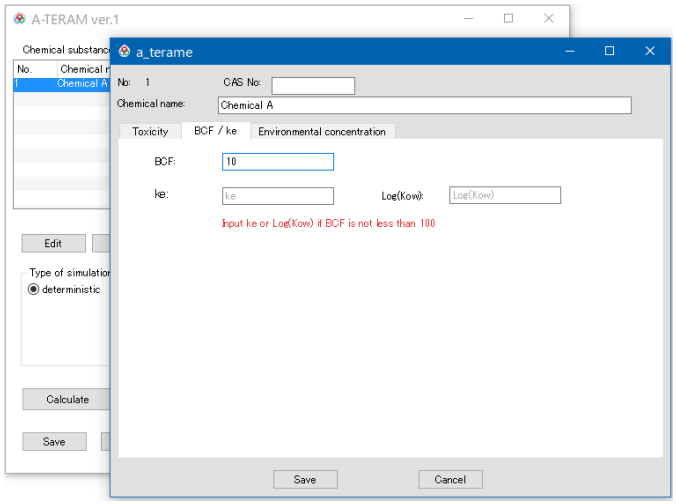


Figure 1- 4 “BCF/ke” sheet in the chemical substance file

After inputting all available ecotoxicity data, select the “BCF/ke” sheet and enter the bioconcentration factor (BCF) for the fish (see Figure 1-4).

If BCF is equal to or larger than 100, enter the elimination constant ke or log(Kow) (logarithm of octanol-water partitioning coefficient). If both data are available and input, the elimination constant has the priority.

If there is no available data on bioaccumulation at all, or if you want to ignore the potential effect of bioaccumulation which might affect long-term toxicity, assign an arbitrary value less than 100 to BCF. If the BCF is less than 100, A-TERAM sets the default value for the elimination constant and outputs the simulation result assuming no bioaccumulation by the chemical in the fish body.

1 -5 Setting of environmental concentration

Setting or inputting data of toxicant concentrations in the environment is performed on the environmental concentration sheet of the chemical substance file (see Figure 1-5). To open the environmental concentration sheet, place the cursor on the tag of "Environmental concentration" and click.

Three types of built-in exposure schemes (“constant concentration”, “stationary fluctuating”, and “seasonally changing”) can be set for different temporal patterns of exposure concentrations.

The constant concentration scheme assumes that the concentration in the environment is constant.

The stationary fluctuating scheme assumes that the concentration in the environment fluctuates randomly over time, however the average level across time does not change throughout the year. The input concentration data corresponds to the expected (mean) environmental concentration, MEC. The environmental concentration at each time step is set as a random sampling from a normal distribution with a standard deviation from MEC. The environmental concentration at each time step is set on a daily basis, and it is assumed that there is no autocorrelation between the time steps (independent). When SD is set as a value extremely smaller than 1, the exposure concentration following the stationary concentration scheme becomes almost identical with the constant concentration scheme.

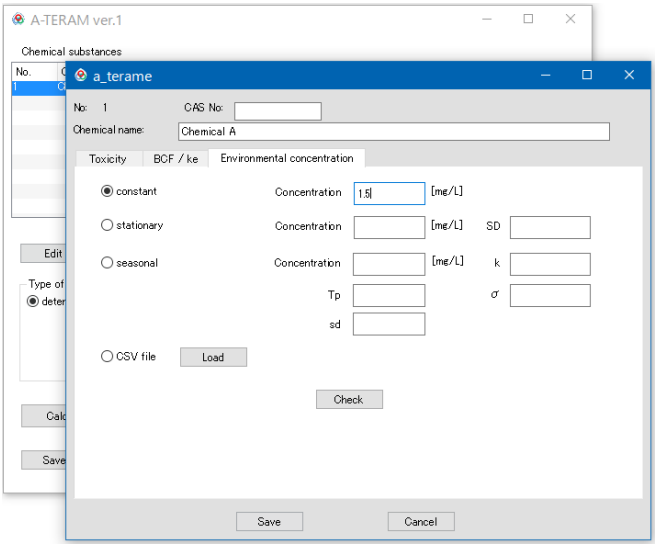


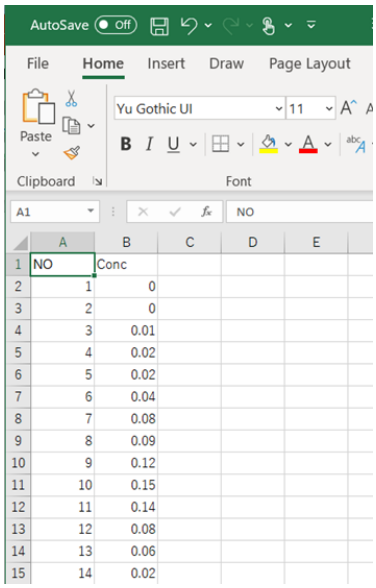
Figure 1-5 Environmental concentration sheet

The seasonally changing scheme assumes that the concentration in the environment fluctuates depending on the season (for details, refer to 2.3.2 “Seasonally changing concentrations in the environment”). It is assumed that the chemical concentration shows a peak-like pattern where the peak of the concentration appears on T_p th day after April 1 which is set as the first day. The chemical concentration you enter is equivalent to the peak concentration. The shape of the concentration pattern with respect to time is determined by two parameters σ and *k* (both take positive values). σ represents the length of the period during which the chemical substance appears in the environment (the larger σ means the longer exposure period), and *k* represents the extent to which the environmental concentration of the chemical is concentrated at the time when the peak concentration appears (the smaller *k* results in the sharper shape of the concentration pattern, indicating that the exposure of chemicals are more concentrated at the peak time).

Furthermore, as in the case of the stationary fluctuating scheme, it is assumed that the environmental concentration at each time step fluctuates randomly with a normal deviation of the variation coefficient *sd* around the mean value of the concentration expected from the exposure pattern. The seasonally changing concentration scheme is relevant for environmental concentrations of chemicals that are expected to be released into the environment in a season-specific manner such as agro-chemicals. Analyses of measured environmental concentrations of some

agro-chemicals suggest that σ = 10 and k = 1.4 for insecticides and herbicides (see S-9 for details).

To directly input environmental concentrations of chemical substances as time-series data, arrange the data in csv format. The valid csv data format for A-TERAM is as follows: In the first column enter the daily (ordinal number) with April 1 as the first day, and in the second column enter the environmental concentration in mg/L. (See Figure 1-6). In the first line enter "NO" in the first column and "Conc" in the second column. The daily can be entered from the first day to the 365th day. Specify consecutive days from the first day, and do not leave an interval. If there is a missing day in the environmental concentration data, perform appropriate interpolation so that there is no loss in the input data. Input of daily and concentration data can be omitted for all days later than the last day you input data. In that case, the environmental concentrations in days later than the last day, where there is no input data, are all recognized as 0.



	A	B
1	NO	Conc
2	1	0
3	2	0
4	3	0.01
5	4	0.02
6	5	0.02
7	6	0.04
8	7	0.08
9	8	0.09
10	9	0.12
11	10	0.15
12	11	0.14
13	12	0.08
14	13	0.06
15	14	0.02

Figure 1-6 csv format for environmental concentrations

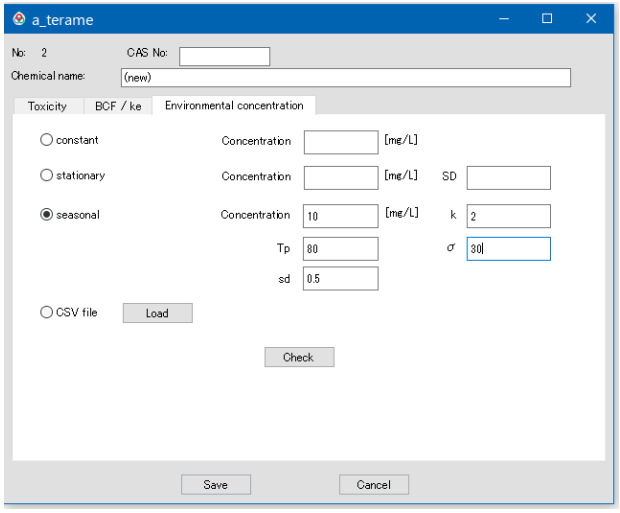


Figure 1-7 Setting of the exposure scheme

To save the csv data, check the csv data on the environmental concentration sheet and click “Read”. Select the csv file that was created in advance and open it.

To confirm the setting or inputting the environmental concentration has been performed correctly, click “Check” for displaying the set values of the environmental concentration on the graph regardless of the concentration schemes (see Figure 1-7). (The horizontal axis is time in days and the vertical axis is concentrations of a chemical substance)

1 - 6 Saving and addition of chemical substances

This completes the inputs of the chemical substance data required for calculating ecological effects. To save these data, highlight the name of the substance on the main screen and click “Save”. A chemical substance information file is created with the chemical substance name as the file name.

To add a new chemical substance to the chemical substance list, click “Add” and repeat the data entry operation. You can add any number of chemicals.

1 - 7　Running a simulation

Save the chemical substance data and return to the main screen to execute the simulation.

A-TERAM can perform three types of numerical calculations: deterministic simulation, stochastic simulation, and com-ECx estimation. In the previous two types of simulations, in each case, the community model consisting of three trophic levels is sequentially calculated from April 1 to April 1 of the following year, and the annual population growth rate λ of the fish, the species at the highest trophic level in the model, is calculated (Fig. 1 - 8). The results of the ecological risk estimation by simulation are denoted as the "ecological risk quotient (ERQ)," which is defined as the rate of decrease in population growth rate under exposure relative to the population growth rate λ_{max} without chemical exposure, 1 - λ / λ_{max} (see Fig. 1 - 9).

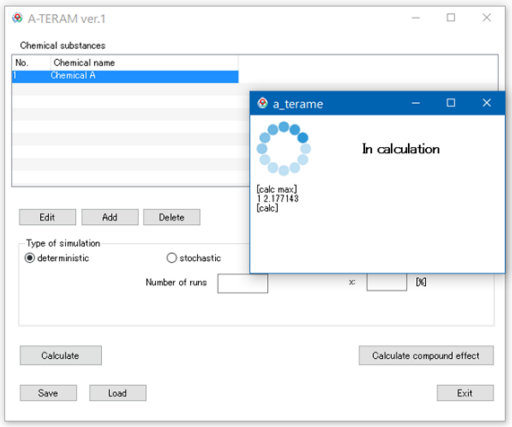


Figure 1-8　A screen panel demonstrating a simulation run

The difference between deterministic and probabilistic simulations lies in the acute chronic extrapolation and whether to account for the uncertainties that arise in estimating the concentration-response function. Deterministic simulations perform calculations using only the mode of the parameters in these statistical estimates, and probabilistic simulations perform calculations that reflect the errors in these estimates. The calculation of the probabilistic simulation is performed by repeating the one-year calculation (one simulation run) many times in order to adopt the parameter values of the acute-chronic extrapolation and the concentration-response function randomly extracted from a specific distribution. The uncertainty in the results of calculation due to these factors can be evaluated and indicated by the

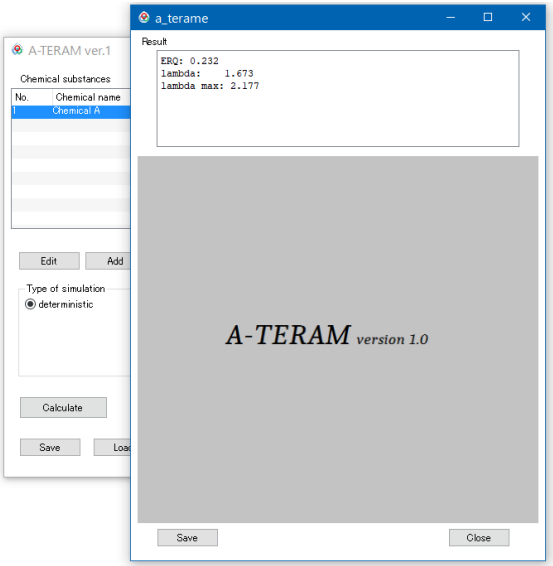


Figure 1-9 The result returned from a deterministic simulation

dispersion of the calculation results.

In the calculation of com-ECx, the chemical concentration at which the population growth rate of the fish decreases by *x* percent is estimated by iterative calculations. The concentration of chemicals is assumed to be constant.

To perform the calculation, select one chemical from the substance list and highlight it in blue (even if there is only one substance in the list, select it). Check the type of calculation to be performed from the “Simulation type” box on the main screen and click “Calculation”. Enter the number of repetitions for stochastic simulation and the effect rate (%) before estimating com-ECx.

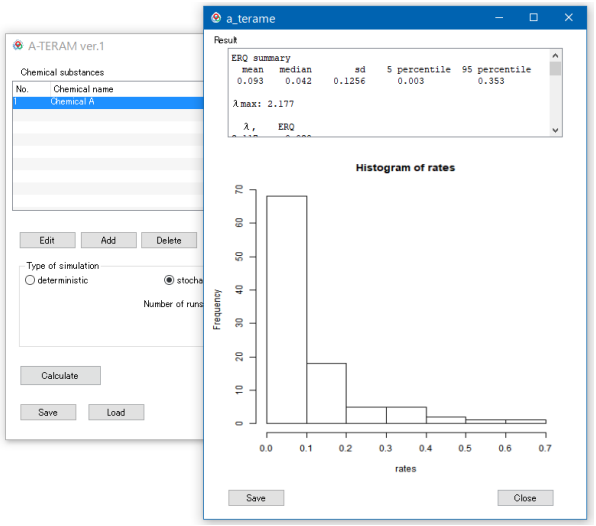


Figure 1 - 10 Results returned from a stochastic simulation

When executing a stochastic simulation, set the number of repetitions considering that one simulation run takes several seconds. To quit the calculation, close the "Calculating" display window by left clicking.

The calculation results are displayed as 5 statistics of ERQ (mean, median, standard deviation, 5th percentile, and 95th percentile), the population growth rate without exposure, and population growth rates of all simulation runs. In addition, histogram of population growth rates is also displayed below the box (Figure 1-10).

Simulation results can be saved as a csv file. Click “Save” and enter the file name and extension (.csv). The result of com-ECx is displayed in unit of mg/L as the concentration of chemicals (see Figure 1-11).

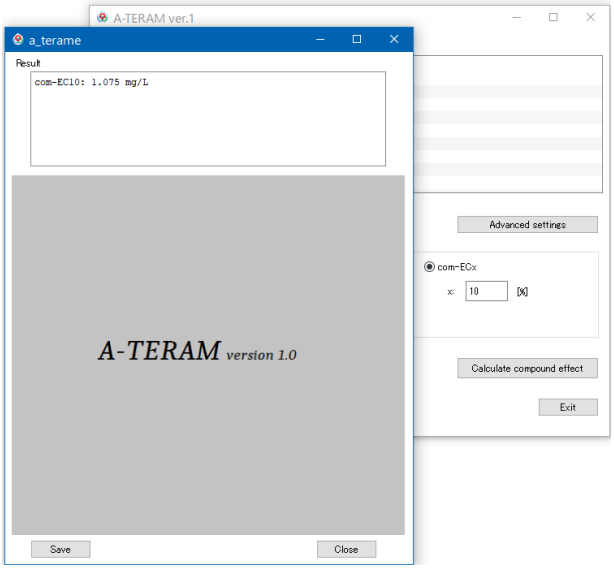


Figure 1 - 11. A display of a com-E10 value

1 - 8 Simulation of mixture effect

A-TERAM can calculate the ecological risk of the mixture effects of multiple chemicals. To assess a mixture effect, two or more chemicals must be listed in the chemical substances file and all necessary data on ecotoxicity, accumulation and environmental concentrations must be entered. To add a chemical to the list, open the main screen, click “Add”, and repeat the data entry procedure described before (see Figure 1-12).

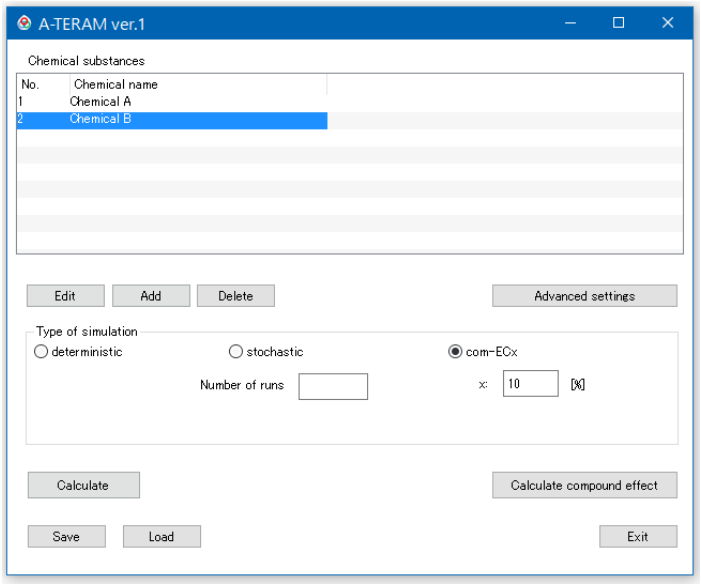


Figure 1-12　Addition of a chemical in the substance file.

Patterns of environmental concentration need not be the same between chemicals. Also, time series data of environmental concentrations can be input in csv format for any component chemicals.

The combined effect is evaluated for all chemicals included in the substance list. For chemicals not to be included as components that cause combined effects, highlight them in blue and click “Delete” to delete them from the chemical substance list.

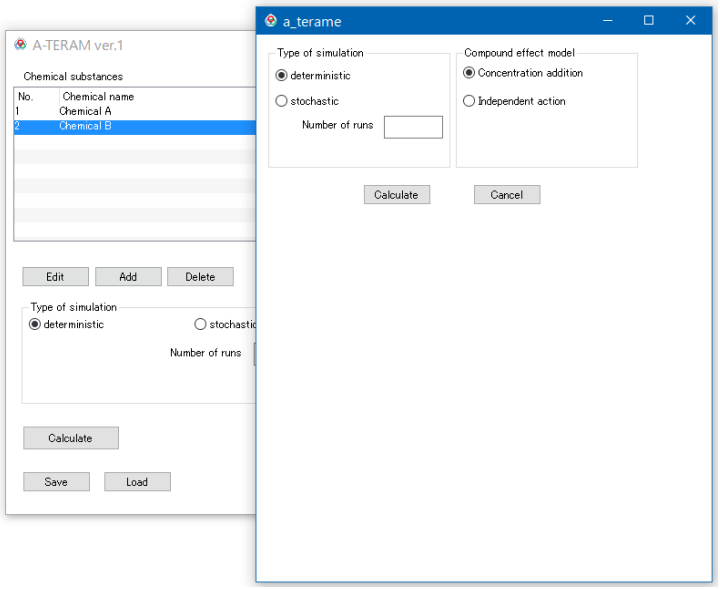


Figure 1 - 13　Control screen for mixture effect simulations

Click “Calculate Mixture Effects” to start simulating mixture effects. The control screen for the mixture effect simulation appears (see Figure 1-13). Mixture effects are predicted using either the "concentration addition model" or the "independent action model" as a reference mixture effect model. Please choose one of them. Either of the mixture effect models can independently apply to all six kinds of responses to chemicals incorporated into A-TERAM. The simulation type is the same as for single substance cases. However, com-ECx cannot be calculated because com-ECx is not defined for mixture effects.

1 - 9　Customization of model parameters

In general, the parameter values of an ecological model represent ecological assumptions, in addition to the species characteristics and environmental conditions assumed. The parameter values in A-TERAM are set based on knowledge of aquatic biology and ecology so as to be consistent with the observation of field populations of medaka fish (*O. latipes*).

However, the values of key parameters can be changed without changing the basic structure of the model, so that the risk can be calculated under ecological assumptions different from the default settings.

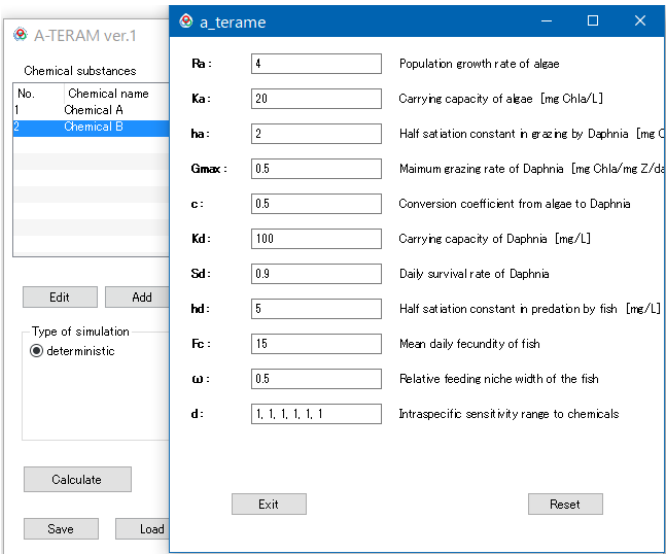


Figure 1-14　Screen for setting model parameters

To change the parameter value, click “Advanced Settings” on the main screen. The parameter list display window appears (see Figure 1-14). Each line describes the parameter symbol, the default value (in the box) and the name of parameter. Enter the parameter value you want to change in the box and click “Close”. To reset all parameter values to the default values, click “Reset to default values”.

Names and meanings of parameters are given below.

Ra："Population growth rate of algae" indicates how many times a major phytoplankton, such as green algae, can grow per day when given sufficient nutrients, water temperature and light intensity. Higher values tend to mitigate the toxic effects on algal growth inhibition.

Ka："Carrying capacity of algae" indicates the maximum biomass of phytoplankton that can grow per unit volume of environmental water. It is an indicator of the density effect of algae (the smaller the value, the greater the density effect).

ha：“Half satiation constant in grazing by Daphnia” means that when Daphnia feeds on algae, the amount of food per Daphnia individual (biomass) saturates against the biomass of the algae (when the algae is increased, the algae will not eat above a certain amount due to satiation). The smaller the value, the greater the amount of food consumed when the density of algae is low, and it means that the food becomes saturated before the algae increases.

Gmax：“Maximum grazing rate of Daphnia” indicates the maximum amount of algae that a Daphnia eats per individual (unit biomass) when there is sufficient algae to feed. The higher the value, the less the ecological effects through acute and chronic effects of Daphnia. Conversely, the lower the value, the greater the ecological effects through acute and chronic effects of Daphnia, and the less the ecological effects of chemicals through algal growth inhibition.

c："Conversion coefficient from algae to Daphnia" means the efficiency of the biomass of algae fed by Daphnia being assimilated into Daphnia individuals and converted to the next generation biomass of Daphnia by reproduction. Changes in ecological effects of chemicals caused by changes in values are similar to those of Gmax.

Kd："Carrying capacity of Daphnia refers to the maximum biomass of Daphnia per unit volume of environmental water when there is sufficient food. It is an indicator of the density effect of Daphnia caused by factors other than the deficiency of algae as a bait resource (the smaller the value, the greater the density effect).

Sd：“Daily survival of Daphnia” means the natural survival of Daphnia, excluding food deficiency and death from fish predation. The higher the value, the less the ecological effects through acute and chronic effects of Daphnia.

hd："The half-saturation constant in predation by fish" means that when fish prey on Daphnia, the amount of predation per individual fish (biomass) saturates against the biomass of Daphnia. The smaller the value of h_d, the greater the amount of predation by fish when the number of Daphnia is small. And fish satiates sooner before the abundance of Daphnia becomes large.

ω："Relative feeding niche width of the fish" indicates the degree to which fish can use various foods other than Daphnia as necessary food resources for reproduction. When $\omega = 1$, Daphnia is not required for fish reproduction, and the ecological effects of chemicals via algae and Daphnia will be assessed regardless of the magnitude of the direct toxic effects on these species. Conversely, when $\omega = 0$, fish cannot breed without Daphnia, and the ecological effects of algae and Daphnia through interspecific interactions are most highly evaluated.

d："Intraspecific sensitivity range to chemicals" indicates how much the sensitivity (response threshold) of adverse responses to a chemical substance varies within a species. The variation range in which the response threshold for a chemical substance differs between individuals is expressed on a logarithmic scale of the concentration of the chemical. The greater the sensitivity range, the smoother the shape of the concentration-response curve at the population level. The default settings assume that one, or susceptible, intraspecific

variation is one order of magnitude. Six types of toxic reactions (fish acute toxicity, fish growth inhibition, fish reproduction inhibition, Daphnia acute toxicity, Daphnia reproduction inhibition, and algal growth inhibition) can be individually set.

Details in A-TERAM including model structure, assumptions, parameterization, sensitivity analyses, and submodel description are also given in Tanaka, Y. et al. (2020) “A 3-species aquatic community model for ecological risk assessment using basic ecotoxicity data” *Environmental Toxicology and Chemistry* 39: 1086 – 1100 and its supporting information.

Part II　Guidance

A-TERAM defines time t as the number of days with April 1st as the first day and simulates the daily change in biomass (population) of organisms at three trophic levels for one year (t = 1-365). By the simulation, the rate of decrease in the annual population growth rate of the top species is calculated is evaluated as "ecological risk quotient, ERQ".

In the guidance, the structure of the A-TERAM model, the meaning and the basis of parameter values, the method of using the input data, ecological models, individual growth model, toxico-kinetics of chemical substances, toxic response model and toxic data analysis method, the environmental concentration of the substance are explained.

2. 1 Dynamics of Biotic Communities

2. 1. 1 Assumed ecosystem

A-TERAM envisions small-scale or semi-static freshwater ecosystems such as ponds and marshes, agricultural drains, and coastal lakes in temperate zones where small freshwater fish inhabit. We also assume a eutrophic and productive environment in which the growth of algae is not limited by lack of sunlight or nutrients. Such an environment is not representative of a variety of freshwater bodies, including the upper and middle watersheds of rivers and offshores of lakes and marshes, but is representative of many rural and suburban waters, and is relevant for ecological risk assessment of chemicals. It is considered to have generality as the target model ecosystem.

2. 1. 2 Definition of abundances at each trophic level

Numerical or biomass abundances are defined for each trophic level as following.

Algae: Chlorophyll weight per unit water volume (1 liter) in the environment (μg/L)

Daphnia: Total dry body weight per unit water volume (1 liter) in the environment (mg/L)

Fish: Number of individuals per unit water volume (1 m^3) in the environment (ind/ m^3)

Hence, variables denoting biomass or numerical abundances are

$A(t)$： Chlorophyll density at time t (μgChla/L)

$D(t)$： Biomass density at time t (mg[dry biomass]/L)

$F(t,a)$： Fish numerical density at time t (ind/m^3)

The fish population has subdivided age structure with age in days (a).

2. 1. 3 Population Dynamics of Algae

Algal biomass increases with growth and decreases with feeding by *Daphnia*. The biomass of algae consumed by *Daphnia* per unit biomass follows the Holling II equation (Bonsall and Hassell 2007).

$$G_{max}\frac{A(t)}{h_a+A(t)} \quad (1)$$

in which G_{max} is maximum grazing rate of Daphnia, and h_a is half satiation constant in Daphnia grazing of algae.

When the algal population growth rate is written as R_a and the algal carrying capacity as K_a, and the Ricker model is used to simulate the population growth (Yodzis 1989), the algae that increase the population change of algae due to proliferation and decrease by feeding is expressed by the following equation ("*ln*" is natural logarithm).

$$A(t+1) = R_a e^{-\frac{lnR_a}{K_a}A(t)} A(t) - G_{max}\frac{D(t)}{h_a+A(t)}A(t) \quad (2)$$

2. 1. 4 Population Dynamics of *Daphnia*

Daphnia can grow, proliferate, and increase biomass by feeding on algae, but it decreases at a constant rate due to death in starvation. In addition, many species of the genus *Daphnia* and closely related cladocerans tend to appear in the field during a limited period peculiar to the species (Hanazato 1998; Horn and Goldman 1994). A seasonal function that reflects the change is introduced.

The population dynamics of *Daphnia* are expressed by the following difference equation.

$$D(t+1) = \left\{S_d + cG_{max}\frac{A(t)}{h_a+A(t)}10^{-\left(\frac{t-T_{opt}}{T_w}\right)^2}\left(1-\frac{D(t)}{K_d}\right)\right\}D(t) \quad (3)$$

The first term in brackets indicates survival, and the second term indicates the rate at which *Daphnia* per unit biomass contributes to *Daphnia* biomass in the next day by breeding. Here, S_d is the daily survival rate of *Daphnia* and K_d is the environmental carrying capacity (mg/L) of *Daphnia*. *c* is the conversion coefficient from algae to *Daphnia*. The *Daphnia* that consumes the algae at the highest feeding rate, when the environmental condition is optimal, increases the biomass of the Daphnia that increases through individual growth and reproduction. The conversion coefficient represents the ratio of the algae consumed to the increase of *Daphnia* biomass. The higher the conversion coefficient c, the higher the energy efficiency and fertility of *Daphnia*.

In general, many species of *Daphnia* (Crustacea: Brachypoda) occur in limited seasons in the natural environment. In particular, many species such as the genus *Daphnia*, *CerioDaphnia*, and *Moina*, which represent large Cladocerans important in the food chain of freshwater ecosystems, are predominant in spring in lakes and marshes in temperate lowlands and are subject to water temperature and food conditions. It has been known that the number of individuals decreases after spring when it becomes unfavorable in terms of dissolved oxygen and other factors (Hall 1964; Clark and Carter 1974; Jones et al 1979; Hanazato 1992).

The seasonal function $10^{-\left(\frac{t-T_{opt}}{T_w}\right)^2}$ is multiplied to the population growth rate of *Daphnia* by reproduction in order to reflect the seasonality in the dynamics of *Daphnia* in the model. Here, T_{opt} is the optimal date for Daphnia growth, and Tw is the width of the Daphnia growth period. From the results of the field survey in the Kanto area (around Kasumigaura Lake), we set T_{opt} = 30 and T_w = 60, referring to the fact that large daphnids peaked in late April and hardly appeared in late June.

The effect of fish predation to the Daphnia population (top-down effect) is not considered in A-TERAM. The assumption that the fish that prey on *Daphnia* would not have a top-down effect on the Daphnia population is unacceptable in any ecological models. It has been evident in many field studies that

the predator-prey relationship reduces the prey abundance, and the biomass and species composition of the zooplankton community largely depends on the presence or absence of the predator plankton fish (Carpenter and Kitchell 1993).

However, the top-down effect of fish on *Daphnia* largely depends on the fish population density, which depends on the setting of the initial fish population on the model. It also depends heavily on the assumption of the maximum predation per fish individual. Equilibrium populations achieved by ecological interactions with *Daphnia* cannot be assessed, and the top-down effect of fish on *Daphnia* depends only on model assumptions (including parameter settings and initial conditions).

The toxic effects of chemicals acting on *Daphnia* populations are assessed as effects on fish populations through a bottom-up effect through the attenuation of *Daphnia* populations (the effect of a lack of *Daphnia* as a bait deteriorating fish populations). A-TERAM does not consider the situation where the ecological top-down effect of fish to *Daphnia* (the effect of fish predation to reduce *Daphnia*) would mitigate the effect of chemicals through the bottom-up effect from *Daphnia* to fish. In a situation where fish are starving because they have exhausted the *Daphnia* population, the direct toxicity of chemical to the fish might restore the *Daphnia* population (reduced top-down effect) and improve the food limitation for the fish. However, A-TERAM does not evaluate the effect that the ecotoxicity of a chemical would compensate the population-level effect by the community-level effect. The purpose of A-TERAM is limited to unifying the ecological effects of chemical substances on the basis of cascading effects to higher trophic levels through interspecies interaction.

2. 1. 5 Population Dynamics of Fish

The natural population inhabiting in the vicinity of Kasumigaura Lake (Ibaraki, Japan) of medaka fish (*Oryzias latipes*) were presumed as a model population.

The reproductive season of fish is assumed to be limited from the 22nd day (April 22) until the71st day (June 10). Such a setting of reproductive season is consistent with observations in medaka field populations. The lifespan of fish was assumed to be 420 days. Therefore, a = 1 ... 420 (see Figure 2-1).

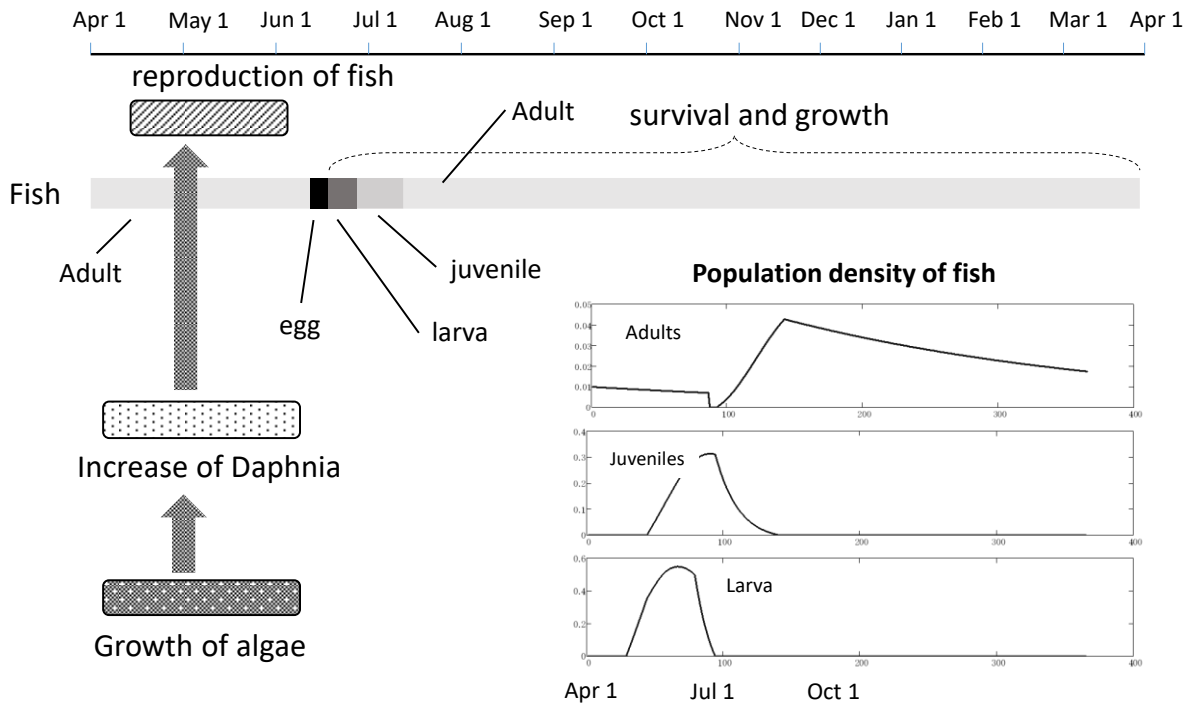


Figure 2-1 Seasonal regimes of reproduction of the 3 species in A-TERAM

Assume that the fish breed (spawn) relying on *Daphnia* and other diets, and that the hatched fry grows with a certain survival rate. The biomass of *Daphnia* eaten per fish was presumed to follow the Holling II equation (Bonsall and Hassell 2007).

Then, the number of eggs laid at time t in the entire population is

$$F(t+1,1) = \left\{\omega + (1-\omega)\frac{D(t)}{h_d + D(t)}\right\} \sum_{a=71}^{420} F(t,a)R(t,a) \quad , \qquad (4)$$

in which h_d is the half-satiation constant for the predation by fish, $R(t,a)$ is per capita reproductive potential of fish with age a at time t (the maximum daily fecundity when foods are enough and there is no exposure to a chemical), and ω is feeding niche width of fish, which means food availability, for the fish, of food items except for *Daphnia*. When ω=1, the fish does not require *Daphnia* for their reproduction, whereas when ω=0, fish cannot reproduce without *Daphnia* and the ecological effect of *Daphnia* and algae through interspecific interaction is most highly evaluated.

The derivation of equation (4) is as follows. The amount of predation per fish per day is written as $P(t)$. When there are enough *Daphnia*, fish eat only *Daphnia*, but when *Daphnia* is deficient, they try to supplement the deficiency with other food items. The optimal switching of food items in predation (Teramoto 1997), which preferentially feeds on other foods when *Daphnia* is rare, was not assumed.

Let P_{max} be the maximum predation rate of fish to Daphnia and P^*_{max} be the maximum predation rate for food other than *Daphnia*. It is assumed that the maximum predation rate reflects not only the ease of

catching the food but also its value as a resource (resource quantity and nutritional value). The term $\frac{D(t)}{h_d+D(t)}$ in the Holling type II predation function indicates the degree of satiation. *Daphnia* feeds on other foods depending on the extent to which it is not saturated. If the efficiency of feeding is very high, the amount of food biomass taken by an individual fish on average is expressed by the following equation.

$$P(t) = P_{max}\frac{D(t)}{h_d+D(t)} + P^*_{max}\left(1 - \frac{D(t)}{h_d+D(t)}\right) \quad (5)$$

Here, because the food availability of foods other than Daphnia relative to that of Daphnia is ω , we get $P^*_{max} = \omega P_{max}$. Substituting it into equation (5) gives $P(t) = P_{max}\left\{\omega + (1-\omega)\frac{D(t)}{h_d+D(t)}\right\}$. Assuming that the number of eggs laid per fish increases in proportion to the food consumed, it is $R(t,a)\frac{P(t)}{P_{max}}$ (the reproductive potential multiplied by the ratio of the maximum *Daphnia* biomass that would be eaten to the actual food biomass that is eaten). Therefore, the number of eggs laid per fish per day is as follows.

$$R(t,a)\left\{\omega + (1-\omega)\frac{D(t)}{h_d+D(t)}\right\} \quad (6)$$

Multiplying the fish abundance of each age class by the daily average fecundity at each age denoted by expression (6) and summing up over all age classes gives equation (4).

Fish populations are determined by increases due to reproduction and decreases due to death. The progressive decrease in fish population at each age due to death is expressed by the following equation.

$$F(t+1, a+1) = S_f(a)F(t,a) \quad \text{if } a \geq 1, \quad (7)$$

in which $S_f(a)$ is daily survival rate of the fish at age a. We set $S_f(a)$ for each age categories corresponding to the egg, larval, juvenile, and adult stages (see 2.1.7).

We hypothesized that fish viability did not depend on the abundance of *Daphnia* as a prey. Long-term fish survival depends on survival during the larval and juvenile stages. In the natural environment, these periods correspond to early summer to early autumn, and most *Daphnia* and closely related species disappear from the environment. In addition, a variety of prey organisms other than *Daphnia* (benthic organisms such as chironomids and annelids, larvae of aquatic insects, etc.) appear in ponds and are dominant. It is thought that it is possible for fish to survive and grow by eating them.

2. 1. 6 Body Growth of Fish

To assess the chronic effects of long-term exposure to chemicals in the early life history of fish, the individual growth of the fish (growth of body size) and the effects of the chemicals on it, and the relationship between individual growth and the rate of increase in the fish population, need to be formulated. In A-TERAM, the following von Bertalanffy growth model, the difference equation version, was adopted to model the individual growth of fish (von Bertalanffy 1957; Gurney and Nisbet 1998).

$$L(t+1, a+1) = L(t,a) + \max[\gamma\{L_{max} - L(t,a)\}, 0]$$

where $L(t,a)$ is the body length (mm) of fish of age a at time t, γ is body growth rate of the fish, and L_{max} is the maximum body length (mm).

We assume that body size affects the rate of fish population growth by determining the maximum number of eggs fish laid. In the actual numerical simulation, the body size of the adult fish at the start of the calculation (April 1) is calculated before the simulation of the population assuming that all individuals were born on May 1 in the previous year (all conditions such as the exposure concentration of chemical substance are set to be the same as the present year). The body size dynamics are not affected by the fish population dynamics unless the density effect is considered. Thus, the body size can be calculated separately from the population fluctuations. (For the dynamics of fish body size and the concentration of chemicals in the body, see 2.1.6 “Individual growth of fish” and 2.2.1 “Disposition of chemical substances”).

Since fecundities $R(t,a)$ is known to be approximately proportionate to body volumes or cubic lengths of the fish (Wootton 1979; Roff 1984, 1992), the reproductive potential is assumed to be subject to the following equation,

$$R(t,a) = \begin{cases} R_{max}\left(\frac{L(t,a)}{L_{max}}\right)^3 & if\ L(t,a) \geq L_\alpha \\ 0 & otherwise \end{cases} \quad (8)$$

where L_{max} is the maximum body length, R_{max} is the maximum reproductive potential when there is no exposure to chemicals, and L_α is the body length at the first reproduction.

2. 1. 7 Settings of ecological parameters and the index of ecological risk

The values of ecological parameters set as default values in A-TERAM are shown in the table. These settings are based on literature on life-history characters obtained from literature surveys on freshwater ecology and limnology, and personal observation on the life history of medaka obtained from field surveys in paddy fields in the vicinity of Kasumigaura Lake. For details on the settings of parameter values, refer to Supporting Information: S.5 *The conjectured ecological parameters*.

Symbol	Name and description	Assigned values (units)
t	Time in days for a year starting from April 1	*var* (1)(days)
$A(t)$	Chlorophyll density (the algal biomass)(2)	*var* (μgChla/L)
$D(t)$	Biomass density of *Daphnia* (dry weight)(3)	*var* (mg/L)
$F(t, a)$	Numerical abundance of the fish (individual numbers)	*var* (ind/m^3)
R_a	Population growth rate of algae	4 (per day)
K_a	Carrying capacity of algae	20 (μg Chla/L)
G_{max}	Maximum grazing rate of *Daphnia*	0.5 (μg Chla/mg Z/day)(4)

c	Conversion coefficient from algae to *Daphnia*	0.5 (μg Chla/mg Z/day)
h_a	Half-satiation constant in grazing by *Daphnia*	2 (μg Chla/L)
K_d	Carrying capacity of *Daphnia*	100 (mg/L)
T_{opt}	Optimal timing of the reproduction period for *Daphnia*	30 (day)
T_w	Width of the reproduction period for *Daphnia*	60 (day)
S_d	Daily survival rate of *Daphnia*	0.9 (per day)
h_d	Half-satiation constant for predation by the fish	5 (mg/L)
F_c	Maximum per-capita daily fecundity of the fish	15 (eggs)
$S_f(a)$	Daily survival rate of the fish with age a	0.94 [$1 \le a \le 70$][5] 0.996 [$71 \le a \le 420$][6]
$L(t, a)$	Body length at time t with age a of the fish	*var* (mm)
L_{max}	Asymptotic maximum body length of the fish	29 (mm)
L_α	Body length at the first reproduction of the fish	20 (mm)
γ	Body growth rate of the fish	0.00914 (rate per day)
C_L	Coefficient in the body growth function	*estim* [7][8] (unitless)
L_b	Body length at hatching of the fish	2 (mm)
ω	Feeding niche width of the fish	0.25 (proportion)
$R(t,a)$	Per-capita reproductive potential of fish of age a at time t	*func* (per female per day)[9][10]
α_m	age of maturity	71 (days)
α_{max}	Maximum age (lifespan)	420 (days)

Notes: (1) *var* denotes a variable, (2) the initial value was set $A(1)=10$, (3) the initial value was set $D(1)=0.5$, (4) micrograms of chlorophyll biomass per milligram of dry *Daphnia* biomass per day, (5) premature period, (6) mature period, (7) *estim* denotes values to be estimated for each vital property, species and chemical, (8) this coefficient was used for determining the body growth rate γ from data, but not used for the simulation model, (9) *func* denotes a function, (10) average numbers of eggs laid by a parental female per day.

The indicator of ecological risk assessed by A-TERAM is the rate of decrease in the population growth rate per year of the fish, the species at the highest trophic level, and is called the ecological risk quotient (ERQ). The population growth rate is defined as the ratio of the number of individuals in the following year to the number of individuals in the previous year, N^*/N (N is the number of individuals in one year and N^* is the number of individuals in the next year). Therefore, the population increases when the population growth rate is greater than 1, while decreases when it is less than 1. When it is 1, the population does not increase or decrease.

The annual population growth rate of fish is defined as the ratio of the adult population on April 1 one year later to the adult population on April 1 (at the start of the simulation) in the model, which is given by

$$\lambda = \frac{\sum_{k=\alpha_m}^{\alpha max} F(365,k)}{F(1,335)} ,$$

where F (1,335) is the initial population size (the simulation assumes that all individuals start from adult fish born on May 1 of the previous year, i.e., individuals at 335 days of age), and α_m is number of days needed for sexual maturity. Denoting the population growth rate without exposure to a chemical as λ_{max} and the population growth rate with exposure to a chemical as λ^*, ERQ is defined as

$$\mathrm{ERQ} = 1 - \frac{\lambda^*}{\lambda_{\max}} .$$

2. 2 Toxic Responses to Chemicals

2. 2. 1 Toxico-kinetics in fish body

For fish that have a long lifespan and are relatively easy to obtain information on bioaccumulation, A-TERAM models the toxico-kinetics of chemical substances and uses the toxicity information obtained from short-term toxicity tests to extrapolate into toxic responses over a long period of time.

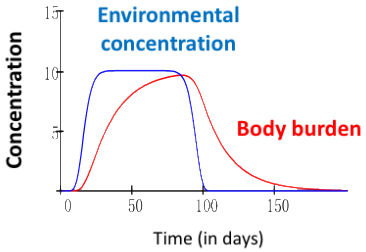


Figure 2-2. Changes in concentrations of a chemical in the environment and fish body

The environmental concentration at time t is $X(t)$ (mg / L), and the body burden (concentration in the body) is $C(t)$ (mg / L). From the first-order toxico-kinetics model, assuming that the uptake constant of a chemical substance is k_i and the elimination constant of a chemical substance is k_e, the change in concentration in the body per unit time is $C(t+1) - C(t) = k_i X(t) - k_e C(t)$ (see Fig. 2-2). It is rewritten as

$$C(t+1) = k_i X(t) + (1 - k_e) C(t) \qquad (9)$$

(Newman and Clements 2008). The bioconcentration factor BCF is equivalent to k_i / k_e . Scaling the body burden in terms of BCF, we get $C^*(t) = \frac{k_e}{k_i} C(t)$. C^* is the scaled concentration in the body such that it would be equivalent to the environmental concentration when it is actually equal to BCF times higher concentration than the environmental concentration (Newman and Clements 2008)。

By putting $C^*(t) = \frac{k_e}{k_i} C(t)$ into it, equation (9) is rewritten as follows,

$$C^*(t+1) = k_e X(t) + (1 - k_e) C^*(t) \quad (10)$$

In the actual simulation, the time series of the concentration in the body were calculated from the time series data of the concentration in the environment using the following formula,

$$C^*(t) = k_e \sum_{\tau=1}^{t-1} (1 - k_e)^{t-\tau-1} X(\tau) \quad . \quad (11)$$

Thus, the rate at which a chemical reaches its maximum concentration in the body by bioaccumulation is determined solely by the elimination constant k_e. The higher the elimination constant, the faster the bioaccumulation process proceeds, reaching the maximum concentration in the body in a short time. Conversely, the smaller the emission constant, the slower the bioaccumulation process, and the longer it takes to reach the maximum concentration in the body (see Figure 2.3).

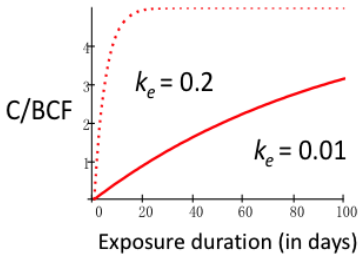


Figure 2.3 Relationship between elimination constants and accumulated body burden

If the emission constant is small, the toxicity may be underestimated when extrapolating the toxicity values estimated in short-term (e.g., 4 days) toxicity tests to the toxicity developed in a long-term exposure environment. Since a typical toxicity test is for a short period of time less than 96 hours, if the elimination constant is small, it is considered that the concentration in the body does not reach the maximum (saturated) concentration in the body or a value close to it during the test. Assuming that the threshold levels in the body that cause a particular toxic response do not change over time, the longer the exposure period, the lower the environmental concentration that causes a particular toxic response. In other words, it is considered that toxicity values such as LC_{50} are time-dependent, and that the longer the exposure period, the lower the toxicity value and the stronger the toxicity (Kooijman 1981).

A-TERAM calculated extrapolated from short-term toxicity data to long-term toxicity based on a toxico-kinetic model, when the bioconcentration factor is high (BCF is 100 or larger), and there can be a large difference between short-term acute toxicity and long-term chronic toxicity for chemicals that take a long time to accumulate in body.

However, the calculation method adopted here assumes that the target organ is not recovered and the threshold concentration in the body does not change with time. In practice, damage of the target organ may tend to recover over time, and the linkage of a toxico-dynamic model that describes the dynamics of such recovering damage with kinetics will provide a more complete picture of the toxic response over time (TKTD model: toxico-kinetic toxico-dynamic model) (Lee et al. 2002; Ashauer et al. 2007, 2011). However, it is very rare that any information on toxico-kinetics is obtained in the normal framework of ecotoxicity testing, and it can be said that considering only the effects of bioconcentration by long-term exposure with the toxico-kinetic model is a safer approach.

The assigned value of elimination constant for each chemical substance was set based on BCF (actually measured by fish accumulation test) according to the following criteria.
(i) When BCF is less than 100: Set k_e = 0.2. Assuming virtually no bioaccumulation, the estimated upper limit (see below) was rounded to one significant digit. In order to maintain continuity with substances with a BCF of 100 or larger, the value was set equal to the estimated upper limit.
(ii) When BCF is 100 or larger: Set an estimated value $k_e \approx 10^{-0.66\log(K_{ow})+0.95}$ based on K_{ow} please refer to S-4). If K_{ow} exceeds the boundary values (log K_{ow} <2.6, log K_{ow}> 6.2), set k_e = 0.17 and k_e = 0.0007, respectively.

2. 2. 2 Toxicity Response Model

The individual traits of an organism that affect its survival and reproduction are called life history traits. In A-TERAM, chemical hazards affect six life-history traits of fish survival, spawning, individual growth, *Daphnia* survival, *Daphnia* reproduction, and algal growth. And as a result, the population parameters that are supposed to change including fish daily survival rate, fish per-capita reproduction rate, individual growth rate, *Daphnia* survival rate, *Daphnia* per-capita reproduction rate, and algal population growth rate.

The quantitative relationship between the exposure level (or concentration in the body) of a chemical substance and the toxic response is represented by a toxic response model and uses the same mathematical expression regardless of differences in species and life history characters.

The toxic response model is based on the exposure concentration: the concentration of the chemical in the environment for algae and *Daphnia*, and the concentration of the chemical in the fish body (scaled by BCF) for fish. These models describe the quantitative effect of the chemical to reduce life history characters. For all concentrations of chemicals mg / L was used as the unit and transformed to the logarithmic scale (x (t) and c (t)) (if the concentration is 0, the original values were set extremely small values).

$$x(t) = log[X(t)]$$
$$c(t) = log[C^*(t)]$$

A function that describes the response at the individual or population level to chemical exposure is called the hazard function. A-TERAM adopted a general unified threshold model as a hazard function (Jager et al. 2011).

Generally, the reason why the toxic response of an organism to a chemical draws a specific concentration-response curve at the level of a population (or a laboratory population) is explained by the individual tolerance model based on the variation of the response threshold for a chemical among individuals (Finney 1947; Newman and McCloskey 2000) or by the the stochastic death model (Bedaux and Kooijman 1994; Widianarko and Van Straalen 1996). The unified threshold model integrates these two principles into one model and succeeds in describing the concentration response relations of chemical substances most generally.

The concentration (in the logarithmic scale) at which an individual exhibits a toxic response to a chemical substance is defined as a response threshold z. The hazard function of an individual with a specific response threshold assumes the following linear function (see Figure 2-4)

$$h_z(x) = \min[1, \eta \max[0, x - z]] \quad \text{(algae and } \textit{Daphnia}\text{)} \qquad (12a)$$

$$h_z(c) = \min[1, \eta \max[0, c - z]] \quad \text{(fish)} \qquad (12b)$$

Here, η is a response slope, and "min" and "max" are functions that select a minimum value and a maximum value in parentheses, respectively. "Algae and *Daphnia*" and "fish" are distinguished from each other in accordance with whether environmental concentrations or body concentrations are used as exposure concentrations of chemical substances. The reason for adopting the linear function as the hazard function at the individual level is that this is the simplest and most general assumption, and almost no information on the nonlinearity of the response (at the shape of the response curve) at the individual level is available from standard toxicity tests.

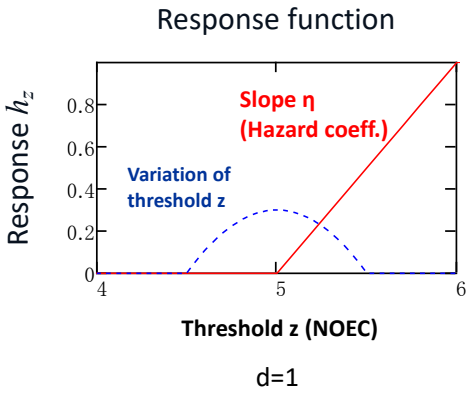


Figure 2-4. Hazard function and distribution of thresholds at the level of individuals

It is assumed that the endpoint estimates (NOEC, EC_{50}, etc.) obtained from toxicity tests reflect the concentration-response relationship when individuals under the experiment consist only of individuals with typical sensitivity (tolerance values) of the species. That is, each toxicity test is performed on a homogeneous strain consisting of only individuals with an average level of response threshold of the species, and the test results are not considered to necessarily reflect intraspecific variations in the response threshold in natural populations.

For algae and *Daphnia*, the mean threshold is determined directly from the no-effect concentration (NOEC) using the following equation:

$$\bar{z} = log(NOEC) \quad \text{(algae and } \textit{Daphnia}\text{)} \qquad (13a)$$

In the case of fish, the concentration in the body accumulated by continuous exposure at the concentration of NOEC is taken as the mean threshold considering the accumulation of chemicals in the body. So, for fish the mean threshold is

$$\bar{z} = log[k_E NOEC \sum_{t=0}^{D-1}(1 - k_E)^t] \ . \quad \text{(fish)} \qquad (13b)$$

Here, D is the test period (exposure days) required for estimating NOEC.

The hazard function at the population level, $\bar{h}(x|\bar{z}, \eta)$, considers the variability of response thresholds among individuals. When the distribution of thresholds in a population is $f(z)$, the hazard function at the population level is

$$\bar{h}(x|\bar{z}, \eta) = \int_{\bar{z}-d/2}^{\bar{z}+d/2} h_z(x) f(z) dz \ , \qquad (14)$$

where d is the range of thresholds in the logarithmic scale. For the case of fish, the value is scaled by the internal concentration of chemicals, and x is replaced by c.

The distribution of thresholds adopts the following quadratic function.

$$f(z) = \begin{cases} \frac{3}{2d}\left(1 - \left(\frac{z-\bar{z}}{d/2}\right)^2\right) & \text{if} \quad |z-\bar{z}| \le \frac{d}{2} \\ 0 & \text{otherwise} \end{cases} \tag{15}$$

With a certain relevant threshold range d, the hazard function at the population level depicts a sigmoid curve (Figure 2-5).

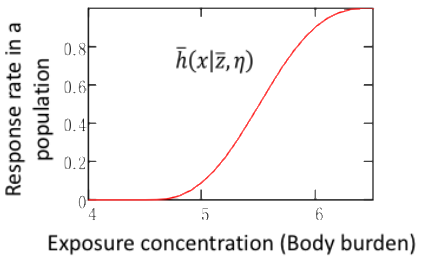


Figure 2-5 Hazard function at the population level

In general, the distribution of sensitivities within species to chemicals is approximated by a (log) normal distribution. A-TERAM adopted the quadratic function instead of the normal distribution as the sensitivity distribution for the following two reasons. Using a normal distribution for a sensitivity distribution results in a model without thresholds for toxic responses at the population level, but there is no basis for using models without thresholds for many chemicals and pesticides. To calculate the population-level toxic response from toxicity and exposure concentration data, it is necessary to perform numerical integration with a normal distribution, and it takes too much time to simulate a community that requires a huge number of calculations (actual calculations of eq[14] with eq[15] used the analytical solution of eq[14], see S-2).

Adopt d = 1 for all life history characters as the default value of the threshold range. In other words, it is assumed that there is a 10-fold (single digit) variation in the response threshold to the chemical within species. This means that toxic reactions are expected to occur in field populations from concentrations as low as about 1/3 ($1/\sqrt{10}$) of the NOEC values obtained in laboratory experiments. The genetic variation of susceptibility in natural populations obtained from a water flea (*Moina macrocopa*) living around Kasumigaura Lake has been shown to be approximately one order of magnitude (see S.3). However, little is known about the degree of variation in chemical resistance (sensitivity) in natural populations of wildlife, which are the true targets of ecological risk assessment.

In A-TERAM, the toxicity of chemicals can cause negative effects on "fish survival (S_M)", "fish growth (L)", "fish fecundity (R)", "Daphnia survival rate (S_D)", "Daphnia reproduction (cG_{max})", and "algal growth (R_a)". The relationship between the toxic effects on the life history characteristics of the species at each trophic level and the population growth at each trophic level is schematically summarized in Figure 2-6.

Changes in ecological parameters (S_M, L, R, S_D, c, R_a) due to each toxic response represented by the hazard function at the population level are expressed by equations (16a) to (16f). An asterisk on an ecological parameter indicates that it reflects a toxic response to the chemical. For fish growth, exposure concentrations are expressed in a time-dependent manner (x_t), and the rate of increase in body length L (body growth rate) is shown as a function of time. It should be noted that the maximum number of eggs laid is also a function of body length (Equation [8]) that elicits indirect impact to reproduction.

① Daily survival rate of fish: $S_M^*(x) = S_M\left(1 - \bar{h}(x|\bar{z}_1, \eta_1)\right)$ (16a)

② Fish growth rate: $L^*(t+1) = L^*(t) + \max\left[\gamma\left\{L_{max} - \frac{L^*(t)}{1-\bar{h}(x_t|\bar{z}_2, \eta_2)}\right\}, 0\right]$ (16b)

③ Fish fecundity: $R^*(x) = R_{max}\left(1 - \bar{h}(x|\bar{z}_3, \eta_3)\right)$ (16c)

④ *Daphnia* daily survival rate: $S_D^*(x) = S_D\left(1 - \bar{h}(x|\bar{z}_4, \eta_4)\right)$ (16d)

⑤ *Daphnia* reproduction: $c^*(x) = c\left(1 - \bar{h}(x|\bar{z}_5, \eta_5)\right)$ (16e)

⑥ Algal growth rate: $R_a^*(x) = R_a^{1-\bar{h}(x|\bar{z}_6, \eta_6)}$ (16f)

In the above equations, $\bar{z}_1 \sim \bar{z}_6$ denote the mean thresholds estimated from the ecotoxicity endpoints of acute fish toxicity, fish growth inhibition, fish reproductive inhibition, *Daphnia* acute toxicity, *Daphnia* reproductive inhibition, and algal growth inhibition, respectively. The methods for calculating the mean threshold values from each toxicity endpoint are summarized in 2.2.6, "Determination of hazard functions based on ecotoxicity input data".

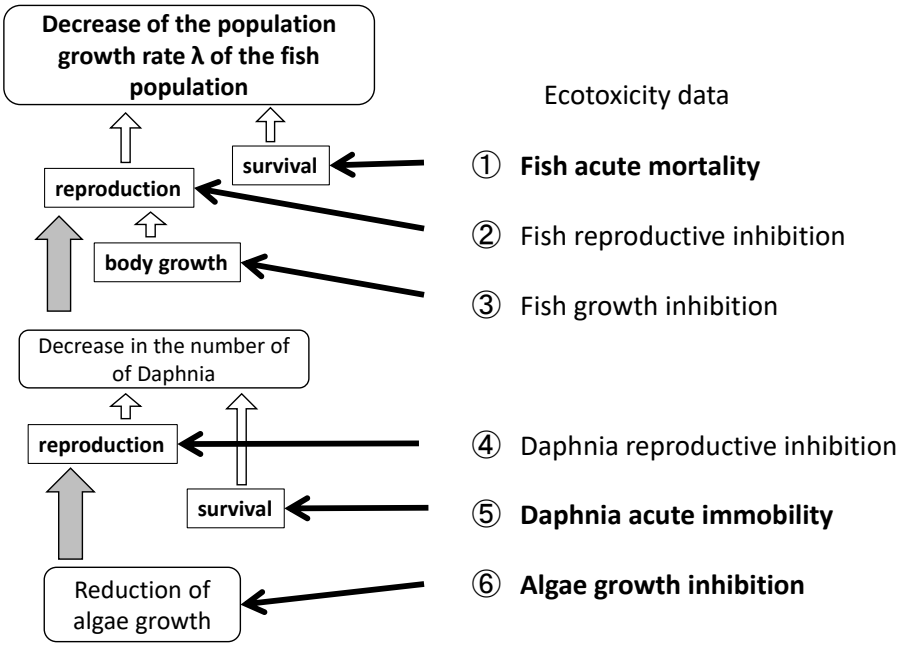


Figure 2-6 Action of ecotoxicities in A-TERAM

2. 2. 3 Empirical distribution of response slopes

The response gradient η is an important parameter that determines the concentration response of ecotoxicity. This section describes how to determine reaction gradients η_1 to η_6.

Regardless of whether the endpoints of ecotoxicity in the input data are NOEC or EC_{50} (or LC_{50}), if the response slopes are different, these endpoints will be used to predict the toxicity response that will be interpolated or extrapolated using the response slope. It makes a big difference. Errors in estimating the response gradient from the toxicity test data may cause significant uncertainty in the predicted value of the toxic response and destabilize the ecological risk assessment results of the chemical.

On the other hand, it is very difficult to obtain a sufficiently accurate response slope estimate from a set of ecotoxicological data, which consist only of several intermediate mortality and immobilization rates between 0 and 1 even if the test is compatible with the OECD Test Guideline and the standard method in the Chemical Substances Control Law. The numbers of treatment of different concentrations are rarely large enough to make the reaction slopes estimation sufficiently accurate. Especially, in fish growth inhibition (early life history) tests and *Daphnia* reproduction inhibition tests that require longer test periods and labor, the number of concentration treatments that are effective for statistical estimation is often further limited.

Therefore, in order to avoid instability in risk assessment due to the uncertainty of the response slope, the response slope of the hazard function for each life history trait is fixed to the most frequently observed value (representative value) for all chemicals (in the case of deterministic simulation). To determine the representative value of the reaction slope, statistical analysis of the toxicity values was performed using the "Ministry of the Environment Ecological Test Results" March 2012 version (hereinafter referred to as "Ministry of the Environment Ecotoxicity Data Base, Ecotox-MoE") as a database. The empirical distribution of the response slope was estimated for ecotoxicities other than fish growth inhibition and reproductive inhibition (see S.6). In this database, both NOEC and EC_{50} (or LC_{50}) are reported for the same test data, and the response slope can be calculated from each individual test data. Regarding the growth inhibition of fish (*O. latipes*), EC_{20} was obtained from reanalysis of the original data (ELS data) of Ecotox-MoE, and the reaction slope was directly estimated from NOEC and EC_{20} (see S.7). As sufficient ecotoxicity data were not available for fish reproductive inhibition, the same value as that for fish growth inhibition was assigned as a representative value of the response slope.

Since the frequency distribution of response slopes was well approximated by the lognormal distribution, the obtained frequency data was fitted to the lognormal distribution (generalized nonlinear regression), and the mode value (the most frequent value) calculated as a parameter of the lognormal distribution estimated by maximum likelihood was adopted as a representative value (see S.6). Fig. 2-7 shows an example of the distribution of reaction slopes (algal growth inhibition).

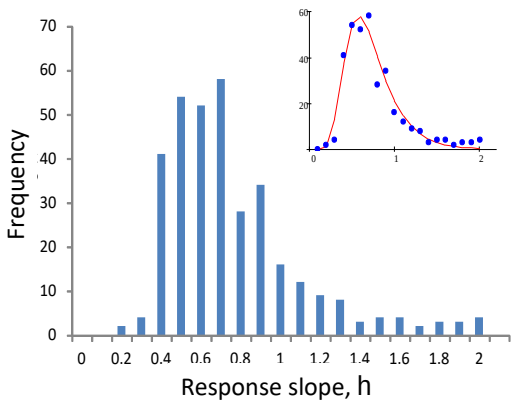


Figure 2-7 Frequency distribution of response slope η in algal growth inhibition. The upper right panel represents the fit of data to the lognormal distribution.

2. 2. 4 Acute-chronic extrapolations

A-TERAM requires fish ecotoxicity (LC50), *Daphnia* acute toxicity (EC50), and algal growth inhibition (NOEC or EC50) as essential ecotoxicity information. If there is no input data for the other three chronic toxicity information, fish growth inhibition (NOEC), fish reproduction inhibition (NOEC), and *Daphnia* breeding inhibition (NOEC), the missing toxicity values are estimated from the required data using extrapolation model. And the estimates are used for calculating the mean threshold in the toxic response model. If there is direct estimate as input data, the direct estimate is used preferentially over the extrapolated estimate.

The extrapolation model is a linear regression model in which essential data are input as explanatory variables and the missing toxicity values are estimated as objective variables. The extrapolation model was created by statistical analysis using ecotoxicity data from Ecotox-MoE as a database of ecotoxicity. In the case of extrapolation between toxicity values, the estimation error is also included in the explanatory variables. Therefore, based on the method of Barnthouse and Suter II (1986), which considers the error of the explanatory variables, the regression equation and the error variance of the objective variable are calculated. In addition, regarding the extrapolation model from the acute immobility to the reproductive inhibition of *D. magna*, it is known that the acute-chronic ratio (ACR) is significantly different between amines and chemicals other than amines (non-amines). They were divided into two groups and extrapolation models were made individually. An extrapolation model could not be created because a sufficient number of effective ecotoxicity values were not obtained for fish reproductive inhibition.

The extrapolation models and the error variances $\mathrm{Var}[\log(\mathrm{NOEC})]$ are listed below. *N* denotes numbers of data (the number of pairs of relevant ecotoxicity endpoints), with which the regression equations were estimated. The unit of toxicity is mg/L for all regression equations.

Fish acute – fish growth inhibition: *N*=29

$$\log(\mathrm{NOEC}) = 0.777\log(\mathrm{LC50}) - 1.17 \tag{17}$$

$$\mathrm{Var}[\log(\mathrm{NOEC})] = 0.219 \cdot \left\{1 + \frac{1}{29} + \left(1 + \frac{0.777^2}{0.604}\right)^2 \cdot \frac{(\log(\mathrm{NOEC}) - 0.044)^2}{97.126}\right\} \tag{18}$$

Daphnia immobility – Daphnia reproductive inhibition:

For non-amines, *N*=255

$$\log(\mathrm{NOEC}) = 0.937\log(\mathrm{EC50}) - 0.961 \quad (19)$$

$$\mathrm{Var}[\log(\mathrm{NOEC})] = 0.262 \cdot \left\{1 + \frac{1}{255} + \left(1 + \frac{0.937^2}{0.878}\right)^2 \cdot \frac{(\log(\mathrm{NOEC}) - 0.673)^2}{1117}\right\} \quad (20)$$

For amines, *N*=55

$$\log(\mathrm{NOEC}) = 1.353\log(\mathrm{EC50}) - 1.739 \quad (21)$$

$$\mathrm{Var}[\log(\mathrm{NOEC})] = 0.522 \cdot \left\{1 + \frac{1}{55} + \left(1 + \frac{1.353^2}{1.831}\right)^2 \cdot \frac{(\log(\mathrm{NOEC}) - 0.663)^2}{145.6}\right\} \quad (22)$$

The acute and the chronic toxicity values used for deriving the above extrapolation models are shown from Figure 2-8 to 2-10.

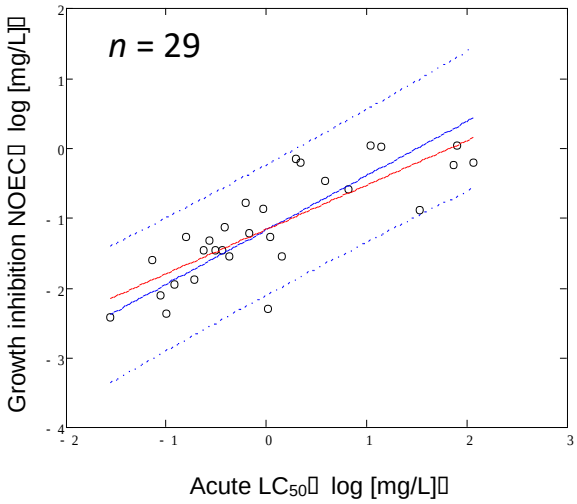


Figure 2-8. Regression of fish growth inhibition NOECs to acute LC50s. The open circles denote observed toxicity values. The blue and the red lines represent the extrapolation model and the usual linear regression model, respectively. *n*: sample size, broken lines: 95 % confidence limit.

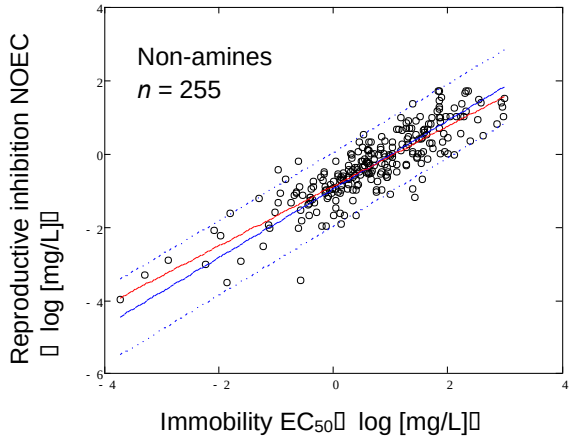


Figure 2-9. Regression of *Daphnia* reproductive inhibition NOECs to *Daphnia* immobility EC50s. The open circles denote observed toxicity values. The blue and the red lines represent the extrapolation model and the usual linear regression model, respectively. *n*: sample size, broken lines: 95 % confidence limit.

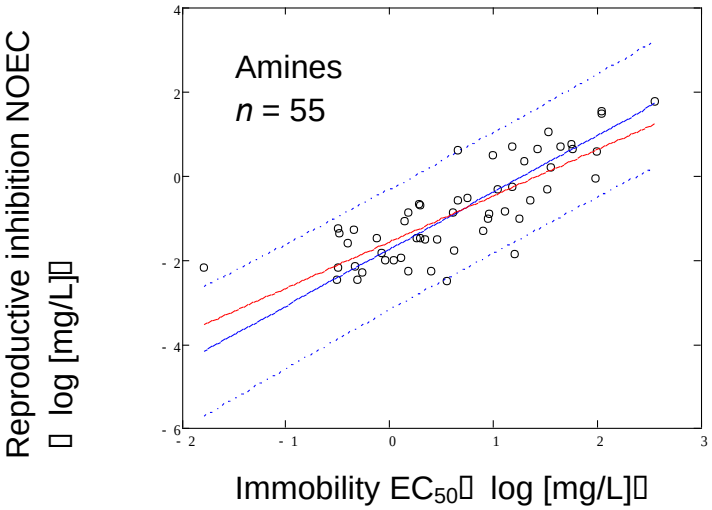


Figure 2-10. Regression of *Daphnia* reproductive inhibition NOECs to *Daphnia* immobility EC50s. The open circles denote observed toxicity values. The blue and the red lines represent the extrapolation model and the usual linear regression model, respectively. *n*: sample size, broken lines: 95 % confidence limit.

2. 2. 5 Deterministic simulation and stochastic simulation

A-TERAM can perform two types of calculations on the input ecotoxicity data: deterministic simulation and stochastic simulation. The difference between the two calculation methods is that as the response slope of the hazard function and the extrapolated value of chronic toxicity it uses the mode (the most frequent value) obtained from existing data as a constant value or a value randomly extracted from the empirical distribution of estimates for each simulation.

In the deterministic simulation, the response slope of the hazard function is fixed as a representative value of the mode of the estimated value calculated from the ecotoxicological data of Ecotox-MoE. When acute chronic extrapolation is performed, the estimate derived from the extrapolation is adopted as the chronic toxicity value. Therefore, for a series of input data, one best estimate is output as the risk evaluation result in the deterministic simulation.

On the other hand, the stochastic simulation outputs the calculation result as a probability distribution that reflects the uncertainty of parameter estimation when calculating the population growth rate and its proportional reduction by pollutants (ERQ, ecological risk quotient). In the stochastic simulation, the response slope of the hazard function for each life history character is determined by random sampling from the empirical distribution (optimally fitted lognormal distribution) of response slopes that was derived from the ecotoxicological data of the Ministry of the Environment, Ecotox-MoE. Furthermore, when acute-chronic extrapolation is performed, a value randomly selected from the lognormal distribution that is based on the expected value and the estimated variance of the extrapolated estimates of log (NOEC) is assigned to the NOEC value in the hazard function. Once these parameter values have been determined, they are fixed in a series of simulations (numerical calculations for community dynamics over a year), but different series of simulations have different random samplings and therefore differ in output results. The frequency distribution of population growth rates λ and ecological risk quotients (decrease rate of population growth rate) can be obtained by a set of many simulations.

The frequency distribution of the population growth rate λ and the ecological risk quotient, ERQ, obtained by the stochastic simulation reflects the uncertain due to estimation of the response slope of the hazard function and the acute chronic extrapolation. Therefore, as more chronic toxicity values are obtained, the uncertainty due to acute chronic extrapolation decreases, and the variability (dispersion) of risk estimation as a decrease in the population growth rate also decreases. A-TERAM does not provide any specific policy or recommendation as to how such variability of risk estimation should be reflected in the risk assessment. Since the deterministic simulation results and the mean or mode of the results obtained from the stochastic simulation are based only on the best estimate of acute-chronic extrapolation, it cannot be said that the uncertainty due to extrapolation is taken into consideration. Defining the 95th percentile of the decreases in population growth rate as an index for ecological risk assessment may be considered to be one of the most effective methods to incorporate such uncertainty of extrapolation into the framework of risk assessment.

2. 2. 6 The determination of hazard functions based on ecotoxicity input data

The procedure for determining the hazard function from the input ecotoxicity information for the six life history characteristics of the three target organisms will be summarized.

① Fish survival rate

Input data：Fish acute LC_{50} (mg/L), test period D (day)

BCF, k_e or K_{ow} (if BCF is larger than 100)

Calculation process：

Step 1

Determine the assigned value of k_e (see S.4).

Step 2

Determine the response slope η.

In the case of deterministic simulation, the mode in the distribution of the response slope of fish acute toxicity (lognormal distribution) in the ecotoxicological data of Ecotox-MoE is adopted.

In the case of stochastic simulation, a value randomly selected from the lognormal distribution is set. Reset the response slope for each simulation. However, in order to prevent the occurrence of exceptional values, the upper limit is set as a value that is twice the standard deviation larger than the average.

Step 3

Calculate the log-transformed value of internal concentration of a substance after the test period, $c_{LC50}(D)$, provided that the exposure test concentration is constantly equal to LC_{50}.

$$c_{LC50}(D) = \log[k_e LC_{50} \sum_{t=1}^{D}(1-k_e)^{t-1}] \quad (23)$$

Step 4

Determine the tentative estimate (guess value) for the mean threshold as $\bar{z} = c_{LC50}(D) - \frac{0.5}{\eta}$.

The mean threshold $\bar{z}$ of the hazard function is determined so as to minimize the deviation from the theoretical value of mortality prediction (50%) by the hazard function (least squares method). $\bar{z}$ is determines so as to minimize

$$f(\bar{z}) = [\prod_{t=1}^{D}\{1-\min[1,\eta\max(0,c_{LC50}(t)-\bar{z})]\}-0.5]^2. \quad (24)$$

② Fish body growth

Input data：fish growth inhibition NOEC (mg/L), the exposure period (pre-match days: D_{pre} and post-hatch days: D_{post})

《not essential》

BCF, k_e or K_{ow} (if BCF is larger than 100)

Calculation process：

Step 1

Determine the assigned value for the elimination constant (see S.4)

Step 2

(i) If NOEC is available,

the mean threshold (logarithmic scale) is determined from the following equation.

$$\bar{z} = log\left[NOEC \cdot k_e \sum_{t=0}^{D_{pre}+D_{post}-1}(1-k_e)^t\right] \quad (25)$$

(ii) If NOEC is not available,

NOEC (mg/L) is estimated by extrapolation from acute LC_{50} (mg/L) (2.2.4 "Acute-chronic extrapolation", Equation [17]), and the mean threshold is determined from equation (25). In the case of probabilistic simulation, the mean threshold is randomly sampled from the lognormal distribution whose standard deviation is set to be equal to the standard error of regression estimation (Equation [18]).

Step 3

Determine the value of response slope η.

In the case of deterministic simulation, the mode in the distribution (lognormal distribution) of the response slopes of fish growth inhibition estimated from the analysis of Ecotox-MoE (see S.6) is adopted.

In the case of stochastic simulation, a value randomly selected from the lognormal distribution is set. Reset the reaction gradient for each simulation. However, in order to prevent the occurrence of exceptional values, the upper limit is a value that is twice the standard deviation larger than the average.

③ Fish reproduction (fecundity)

At present, the data on reproductive ecotoxicity of fish (*O. latipes*) could not be collected sufficiently, and no information was available on the distribution of response slopes and the regression required for acute-chronic extrapolation. Therefore, for the time being, the response slope and the mean threshold in growth toxicity will be applied to reproduction inhibition of fish.

However, in case NOEC is available from reproduction tests, the mean threshold is individually determined with the following equation,

$$\bar{z} = \log[NOEC \cdot k_e \sum_{t=0}^{D-1}(1-k_e)^t] \quad (26)$$

where D is the test period in days.

④ *Daphnia* survival rate

Input data：acute immobility EC_{50} (mg/L), test period D (day).

Calculation process：

Step 1

Determine the value of response slope η.

In the case of deterministic simulation, the modal value in the distribution of response slopes (logarithmic normal distribution) of acute immobility of *Daphnia* in the Ecotox-MoE is adopted.

In the case of stochastic simulation, a value randomly selected from the lognormal distribution is set. The value of response slope is reset for each simulation. However, in order to prevent the occurrence of exceptional values, the upper limit is set as twice the standard deviation larger than the mean value.

Step 2

Determine a guess value of the mean threshold as $\bar{z} = \log(EC_{50}) - \frac{0.5}{\eta}$.

Determine the mean threshold such that deviation of the expected immobility rate given by the hazard function from the theoretical expectation, which is 50 %, by means of the least square method.

$$f(\bar{z}) = [\{1 - \min[1, \eta \max(0, \log(EC_{50}) - \bar{z})]\}^D - 0.5]^2 \quad (27)$$

Minimization of $f(\bar{z})$ predicts the best estimate of $\bar{z}$.

⑤ *Daphnia* reproduction

Input data：Daphnia reproductive inhibition NOEC (mg/L), the test period in days (*D*)

《not essential》

Calculation process：

Step 1

Determine the value of response slope η.

In the case of deterministic simulation, the modal value in the distribution of response slopes (logarithmic normal distribution) of acute immobility of *Daphnia* in the Ecotox-MoE is adopted.

In the case of stochastic simulation, a value randomly selected from the lognormal distribution is set. The value of response slope is reset for each simulation. However, in order to prevent the occurrence of exceptional values, the upper limit is set as twice the standard deviation larger than the mean value.

Step 2

(i) For the case where a NOEC value is available,

The mean threshold (in logarithmic scale) is determined from NOEC (mg/L):

$\bar{z} = \log(NOEC)$. (28)

(ii) For the case where no NOEC value is available,

NOEC (mg/L) value is estimated by the acute-chronic extrapolation using the equation (19) in 2.2.4 "Acute-chronic extrapolation", and the mean threshold is determined from $\bar{z} = \log(NOEC)$.

⑥ Algal growth

Input data：algal growth inhibition NOEC or EC_{50}.

《one of them is essential》

Calculation process:

(i) For the case where only a NOEC value is available,

Step 1

Determine the mean threshold value (in logarithmic scale) from the NOEC.

$\bar{z} = \log(NOEC)$

Step 2

Determine the value of response slope η.

In the case of deterministic simulation, the modal value in the distribution of response slopes (logarithmic normal distribution) of algal growth inhibition in the Ecotox-MoE is adopted.

In the case of stochastic simulation, a value randomly selected from the lognormal distribution is set. The value of response slope is reset for each simulation. However, in order to prevent the occurrence of exceptional values, the upper limit is set as twice the standard deviation larger than the mean value.

(ii) For the case where only an EC_{50} is available,

Step 1

Determine the response slope according to (i) Step 2.

Step 2

Determine the mean threshold based on the test value of EC_{50} and the estimate of response slope from $\bar{z} = \log(EC_{50}) - \frac{0.5}{\eta}$.

(iii) For the case where both NOEC and EC_{50} are available,

Step 1

Determine the mean threshold from $\bar{z} = \log(NOEC)$.

Step 2

Determine the mean threshold from $\eta = \frac{1}{2\{\log(EC_{50}) - \log(NOEC)\}}$. Also, for the case of stochastic simulation, the fixed values are used as the response slope and the mean threshold.

2. 3. Environmental Concentrations of Chemicals

Three typical temporal patterns of concentrations in the environment, "constant concentration scheme", "steady-state fluctuation scheme", and "seasonal variation scheme" are prepared in A-TERAM. The constant concentration scheme means that the exposure concentration is constant throughout the year, the steady-state fluctuation scheme means that the concentration in the environment randomly fluctuates with time, but the average level of concentrations remains constant throughout the year without any temporal trends, and the seasonal variation scheme means that the concentration in the environment changes temporally according to seasons. In the seasonal variation scheme, it is assumed that there is a unimodal pattern that shows the maximum concentration at some time. It is considered in general that industrial chemicals and household/health care products tend to follow the constant or steady-state fluctuating schemes, while agrochemicals such as insecticides and herbicides tend to follow the seasonal variation scheme.

A-TERAM does not include any modules specified for estimating environmental exposure concentrations of chemicals. However, it can input arbitrary time-series data of environmental concentrations besides the three typical exposure schemes and can return risk estimation results (1.5 ”Setting of Environmental Concentrations”).

2. 3. 1 The steady-state fluctuating scheme

The entered environmental concentration is MEC (mg / L). The daily time-series environmental concentration x (mg / L) is created by repeating random sampling from the normal distribution with mean MEC and standard deviation MEC×sd (standard deviation). It is assumed that there is no autocorrelation (concentrations tend to be similar when they are closely related in time) in time-series data.

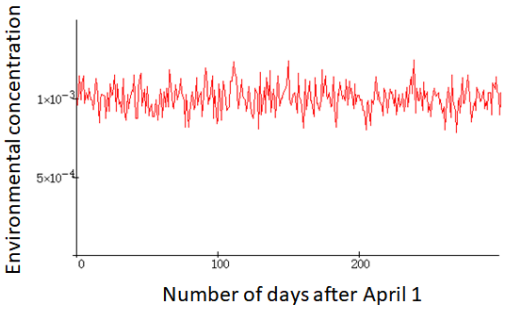


Figure 2-11. Schematic drawing of steady-state fluctuation of environmental concentration

2. 3. 2 The seasonal variation scheme

It is assumed that the environmental concentration (mg/L) exhibits a unimodal seasonal variation that peaks on the T_p-th day (the date of maximum daily concentration). The expected concentration E_x (t) after t

days follows the formula below. The input data of concentrations corresponds to the maximum concentration (the peak concentration) X_{max} in the environment.

$$E_x(t) = X_{max} exp\left[-\left(\frac{|T_p - t|}{\sigma}\right)^k\right] \qquad (29)$$

in which σ is an index for the duration of environmental exposure, k is the skewness of the variability of concentrations along time (S.9).

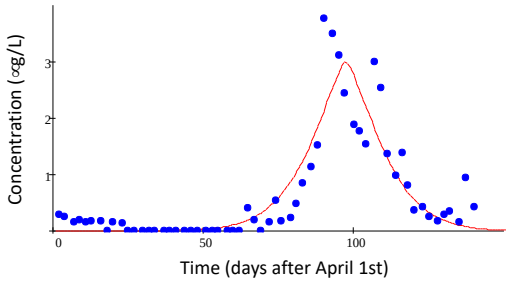


Figure 2-12. An example of seasonal variation scheme (Iprobenphos measured at Kokai river, Ibaraki, Japan)

Considering stochasticity of exposure concentrations, the exposure concentrations (mg/L) are simulated using the below equation,

$$P_x(t) = \max\left[0, X_{max}\left(exp\left[-\left(\frac{|T_p - t|}{\sigma}\right)^k\right] + \varepsilon \sim N(x|0, sd)\right)\right], \qquad (30)$$

where $\varepsilon \sim N(x|0, sd)$ is random sampling errors from the standard normal distribution (white noises) and *sd* indicates magnitudes of random variation in exposure concentrations.

2.4 Discussion and Future Scope

A-TERAM was developed for the purpose of supporting ecological risk assessment with higher ecological relevance by introducing the ecological traits of the species at each trophic level and the ecological interactions, e.g., prey-predator relationship. In this section, we overview results of the analysis that examined the effect of ecological parameters to the risk assessment by A-TERAM and discuss the relationship between the model outputs and the extinction risk of populations or species.

2. 4. 1 Differential relationships between concentrations and population growth rates according to the kind of ecotoxicity

A-TERAM evaluates ecological risks in terms of reductions in the population growth rate at the highest trophic level (Ecological Risk Quotient) as adverse responses at lower trophic levels are supposed to extend to the highest trophic level. This means that even if concentration-response relation in the same vital property (e.g., the fish acute mortality and the Daphnia immobility) is almost compatible between different trophic levels it does not necessarily result in the same concentration-response relation between the different trophic levels in terms of reductions in the population growth rate. This completely holds for

concentration-response relations between different vital properties within a same trophic level (e.g., the Daphnia immobility and the Daphnia reproductive inhibition). These things imply that the present model transforms the concentration-response relation in a particular vital property at a particular species into a concentration-response relation in the population growth rate as the final criterion of ecological risk depending on ecological properties of the species and the role in ecological interactions. In brief, the same response rates in different vital properties or at different trophic levels are likely to have different impacts to biotic communities or ecosystems.

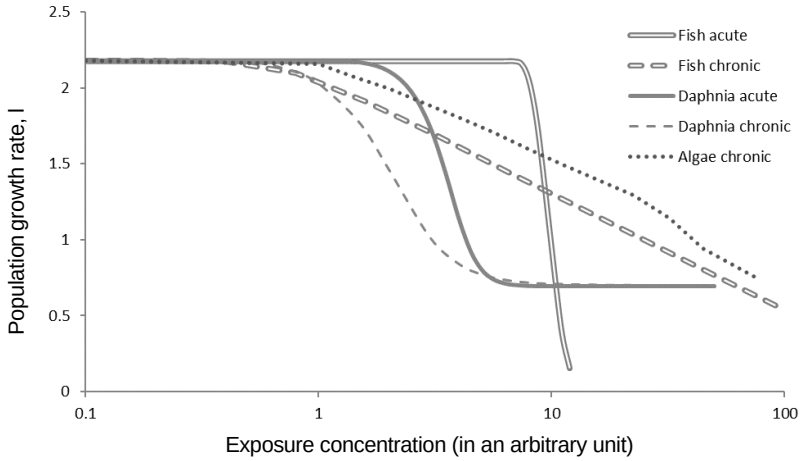


Figure 2.13 Concentration-response relations in fish population growth rate through different kinds of ecotoxicity

In order to understand the characteristics of the quantitative relationship between the ecotoxicity that affects the life history traits at each trophic level and the ecological risk quotient of A-TERAM, the ecotoxicity value was set to 100 for acute toxicity of fish and to acute of Daphnia immobility 10. Chronic toxicity NOEC was hypothetically assigned 1 for all vital properties of all species. And then the concentration-response relationship was calculated when each ecotoxicity acted alone (for example, when Daphnia acute toxicity was the target, toxicity values were set to extremely high values thereby disregarding toxic effects to algae and fish). The ratio of these assigned acute toxicity values to the NOEC value (1) is approximately equivalent to ACR (acute-chronic ratio), and all toxicities are regarded to induce roughly equal ecological hazard in the first-tier risk assessment.

The concentration-response relationship was less steep and nearly linear for the case where the algal chronic (algae growth inhibition) and the fish chronic (fish growth inhibition) were responsible to the toxic effect (Fig. 2.13). The fish acute toxicity caused an extremely steep concentration-response relation in terms of ERQ. The toxicities on Daphnia regardless of whether it is acute or chronic resulted in intermediate sigmoid concentration-response relationships. A general tendency was that chronic toxicities caused more gradual concentration-response relationships than acute toxicities. As for the fish acute toxicity, only trivial increase of daily mortality for long time can drastically exacerbate yearly survival rate and lead to considerable extinction risk of populations.

2. 4. 2 Sensitivities of ecological risk quotients to ecological parameters

The ecological risk assessment with A-TERAM depends on the ecological characteristics of species and interspecific interactions between trophic levels. If the result of risk assessment is greatly influenced by the ecological parameters that define these, and if it is difficult to set the ecological parameters accurately due to lack of ecological information, the ecological risk assessment based on mechanistic models will be highly uncertain. On the contrary, if the ecological parameters can be set within the range where the risk assessment result is not significantly affected, the high reliability of the risk evaluation method may be expected.

To evaluate uncertainties of model results in terms of reduction of under a particular level of exposure to chemicals, which could be driven by imprecise setting of model parameters, the local sensitivity analysis was practiced for proportional decrements of λ (the ecological risk quotient) against a small perturbation of each of all 9 ecological parameters (the population growth rate of algae, R_a; the maximum grazing rate of Daphnia, G_{max}; the half satiation constant in grazing by Daphnia, h_a; the conversion coefficient from algae to Daphnia, c; the daily survival rate of Daphnia, S_d; the half satiation constant in predation by fish, h_d; the feeding niche width of fish, ω; the maximum per-capita fecundity of fish, F_c; and the daily survival rate of fish, S_f). The relative values of local sensitivity (elasticity) against small deviations of each parameter value were evaluated by increasing and decreasing the parameter values by 5 percent from the baseline values and by detecting the subsequent changes in decrements of λ from the reference value without exposure to chemicals. As for the survival rate of the fish S_f, 0.1 percent changes rather 5 percent were made to assess the elasticities because the population growth rate of the fish was extremely sensitive to the survival rate of the fish. It was assumed that only one of the five biotic responses (the fish acute effect, the fish chronic effect, the Daphnia acute effect, the Daphnia chronic effect, and the algal chronic effect) was directly derived by the exposure. The fish reproduction and growth were assumed to respond in the same way and were both included in the fish chronic effect. The hypothetical exposure level was set such that the adverse response of each biotic response induced approximately 10 percent reduction of λ (ERQ=0.1) before introducing the perturbations of ecological parameters.

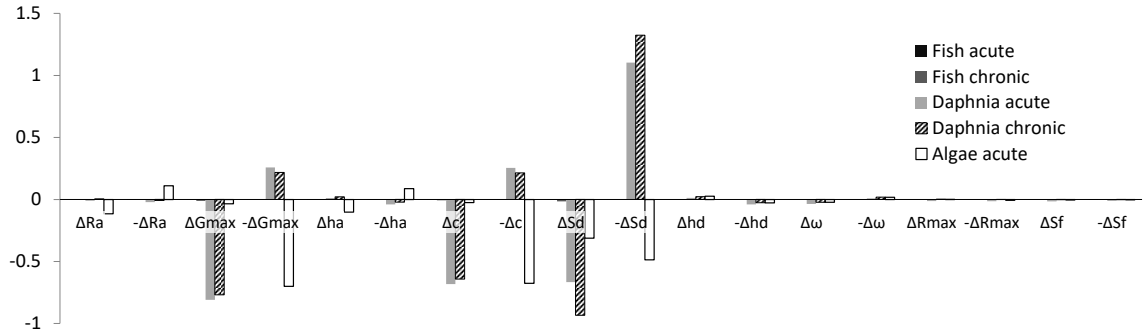


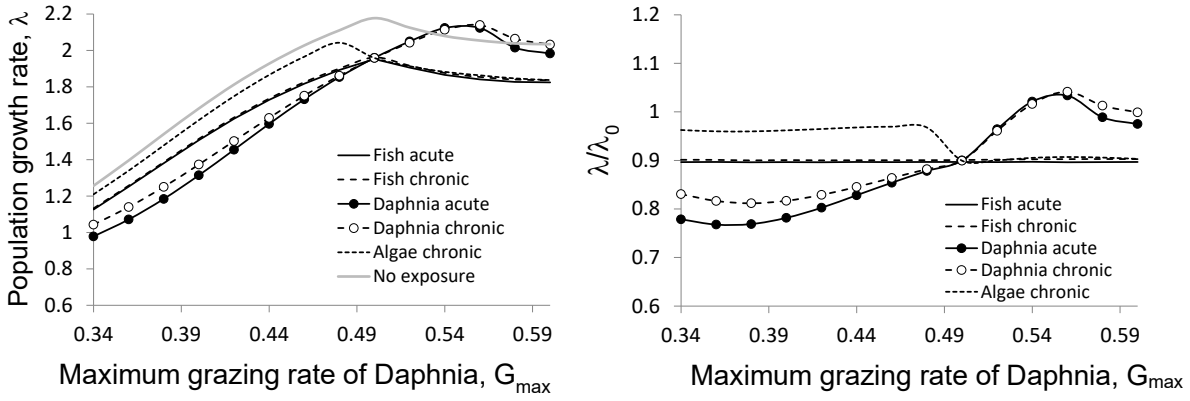
Figure 2.14. Sensitivities of ERQ (ecological risk quotient) against ecological parameters
Changes in ERQs due to 5 percent increase and decrease (Δ or - Δ) of ecological parameters from the default values are indicated. The toxicity values are set following Figure 2.13. The sensitivities were respectively evaluated for each causal ecotoxicity: fish acute, fish chronic (fish growth inhibition), Daphnia acute (immobility), Daphnia chronic (reproduction), and algae acute. The examined ecological parameters are population growth rate of algae (Ra), maximum grazing rate of Daphnia (Gmax), half-satiation constant in grazing by Daphnia (ha), conversion coefficient from algae to Daphnia (c), daily survival rate of Daphnia (Sd), half-satiation constant for predation by the fish (hd), feeding niche width of the fish (ω), maximum daily fecundity of the fish (Rmax), and daily survival rate of the fish (Sf).

Figure 2-14 depicts the elasticities of ERQ in terms of the rate of change in ERQ under deviations of the ecological parameters in comparison to the ERQ under the baseline values of ecological parameters: elasticity = $\frac{\mathrm{ERQ}^{*}-\overline{\mathrm{ERQ}}}{\overline{\mathrm{ERQ}}}$, in which $\overline{\mathrm{ERQ}}$ is the ERQ based on the baseline parameter values, ERQ^{*} is the ERQ under a perturbation of an ecological parameter, and ERQ is commonly defined as $1-\lambda'/\lambda_0$ in each parameter setting (λ' and λ_0 are the annual population growth rate of the fish with and without exposure to a chemical). Thus, the elasticity of 0.5, for example, would indicate that ERQ increased by 50 percent as a result of the small perturbation of an ecological parameter, leaving a warning that the ecological risk estimation based on ERQ in A-TERAM would entail large uncertainties unless the examined model parameter is appropriately determined to assure the ecological reality.

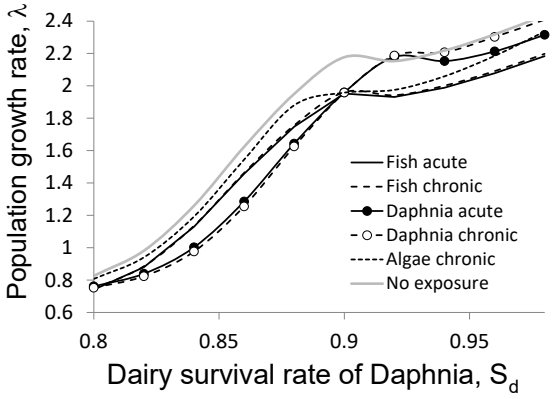
All parameters that influence the ecological characteristics of algae and fish did not significantly affect the results of the ecological risk quotient calculation. On the other hand, many of the parameters that influence the ecological characteristics of *Daphnia* (maximum grazing rate G_{max}, conversion efficiency c, survival rate S_d) showed high sensitivity. This suggests that the setting of ecological parameters related to *Daphnia* must be carried out with particular care for rational risk assessment.

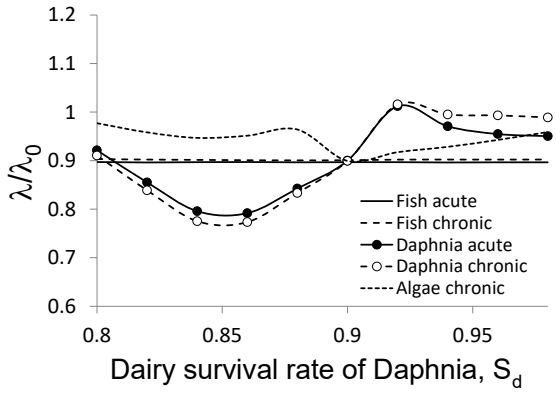
The global behaviors of λ with or without toxicant exposure were also examined (Figure 2.15). Changes of model parameters greatly changed λ with and without the toxicant effect but at nearly the same rate (less than ± 10 percent difference) in all parameters except for the Daphnia traits, G_{max}, S_d and c. These traits when they exceeded the baseline values (say G_{max} = 0.54) greatly diminished the toxicant effect to λ through the acute and chronic effects to Daphnia, because the prospered Daphnia population would have impeded the transmission of the negative impact by pollutants to the Daphnia population up to the fish population. On the contrary, with G_{max} much less than the baseline value, the toxicant effect through the algae growth inhibition almost disappeared, because the algal growth would be enhanced by the reduced grazing pressure by Daphnia so that the negative effect on the algal population was not transmitted up to higher trophic levels. The both extreme conditions do not constitute the reference ecosystem state within the framework of A-TERAM, in which ecological risks are evaluated as a disturbance of a well-balanced ecosystem.

(I) Global properties of λ against changes in parameter values of G_{max}

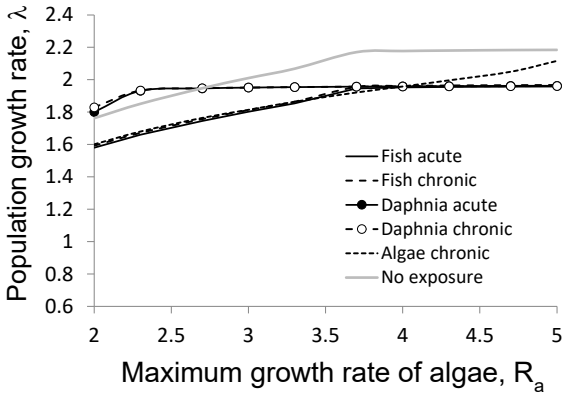


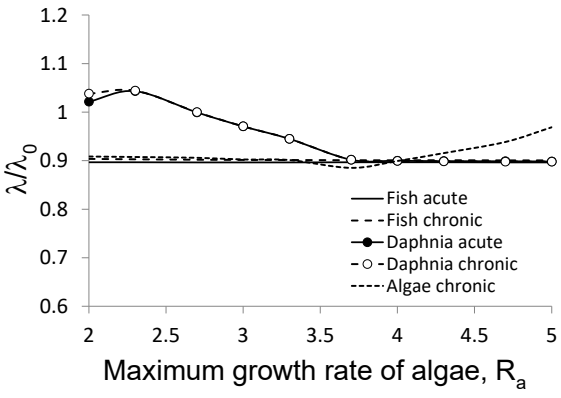
(II) Global properties of λ against changes in parameter values of S_d



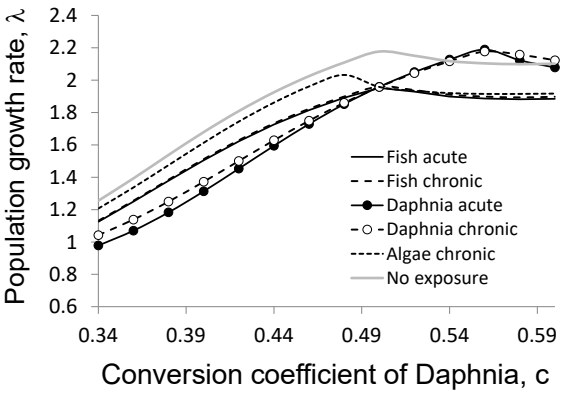


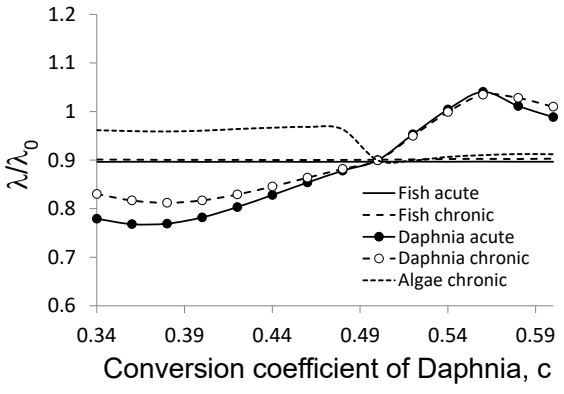
(III) Global properties of λ against changes in parameter values of R_a



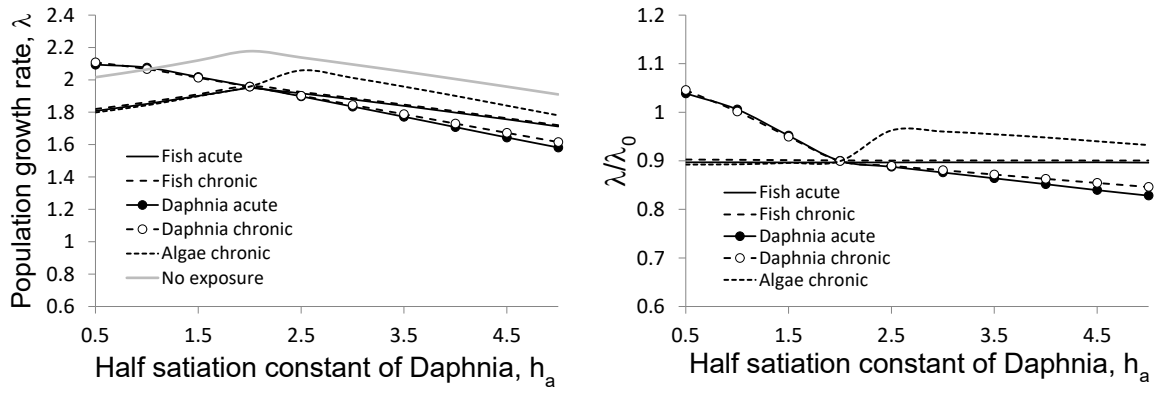


(IV) Global properties of λ against changes in parameter values of c

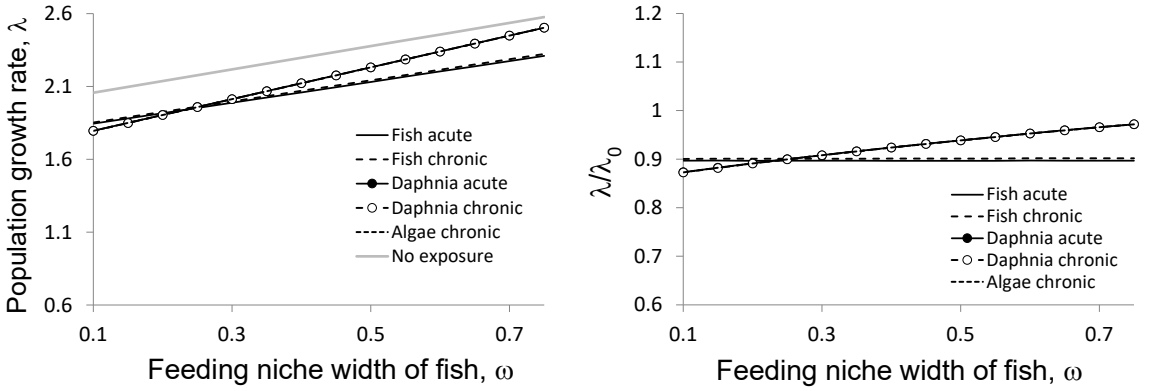




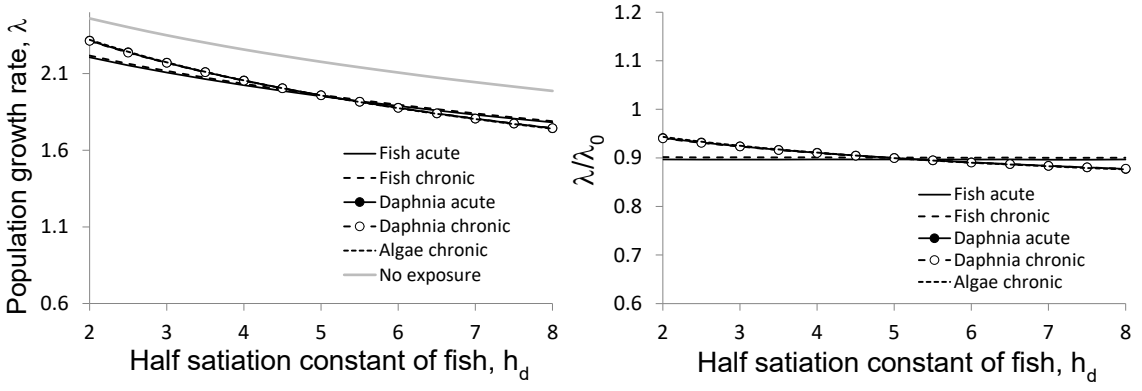
(V) Global properties of λ against changes in parameter values of h_a



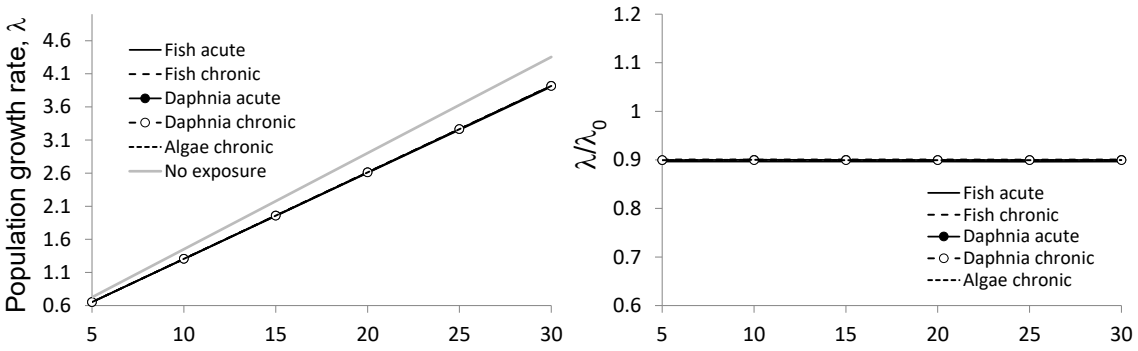
(VI) Global properties of λ against changes in parameter values of ω



(VII) Global properties of λ against changes in parameter values of h_d



(VIII) Global properties of λ against changes in parameter values of F_c



(VIIII) Global properties of λ against changes in parameter values of S_f

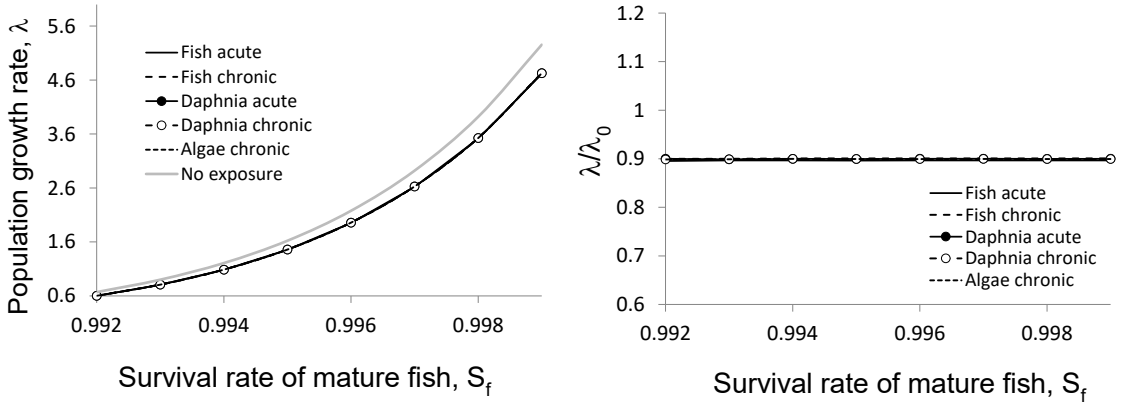


Figure 2.15. Changes in population growth rates λ of the fish responding to each of the 9 ecological parameters. The responses of λ are shown for cases with and without toxicant effect to each vital property. The right panels depict population growth rates of the fish relative to the reference population without exposure (λ_0).

2. 4. 3 Extinction risk: Vulnerability of populations

Population extinction probability is one of the most plausible indicators for quantitatively assessing the effects of various environmental disruptors, including chemical contamination, on wildlife. When the extinction of the target organism is threatened, the extinction of the populations that make up the species can be regarded as a clear ecosystem hazard. Since extinction risk is expressed as the probability that an extinction event will occur, the magnitude of risk can be compared and evaluated as an additive measure for multiple drivers of extinction. Wildlife extinctions are caused by a variety of environmental disruptors other than chemical pollution, including habitat destruction and loss, overhunting, climate change, and the invasion of alien species. By converting the ecological impacts of chemicals into population extinction risks, as well as the various environmental disruptors, it is possible to compare the ecological impacts of chemicals with these environmental disruptors (Tanaka and Nakanishi 2000).

The adverse effects of chemicals on wildlife can be summarized as a decrease in the population growth rate of organisms, as they mainly reduce the fitness of organisms. Population extinction probabilities or mean extinction times ($\bar{T}$) are known to be approximated by the following equation when some assumptions are met (Lande 1993,1998; Lande et al 2003):

$$\bar{T} \propto K^{2E[\ln \bar{\lambda}]/\sigma_e^2} \quad (31)$$

where $\bar{\lambda}$ is the long-term mean population growth rate ("ln" denotes the natural logarithm), K is the carrying capacity, $\sigma_e{}^2$ is the environmental variability of the population growth rate across time, and $E[\ ]$ denotes the expectation. It is assumed here that the population growth rate λ is subject to random fluctuation due to varying environments and the long-term average is $\bar{\lambda}$ and the variance is $\sigma_e{}^2$. The population size is also subject to stochastic variation according to the variability of λ, and the population is regarded as extinct when the population size is expected to decrease below 1.

Assuming that population extinction events are approximately time-independent, the extinction probability p per unit time is equivalent to the inverse of the average extinction time (Tanaka 2003). And the increment of extinction probability Δp due to the chemical exposure is approximated by the following equation (see S-10),

$$\Delta p \cong p_0\left(K^{(2/\sigma_e^2)ERQ} - 1\right) \quad (32)$$

where p_0 is the background value of extinction probability (the background extinction risk), which indicates the extinction risk of focal populations due to any causes except for chemical pollution $p_0>0$)。

The proportional increase of extinction risk relative to the background extinction risk $\Delta p/p_0$ is an exponential function of ERQ ($\Delta p/p_0 = e^{ERQ(2/\sigma_e^2)\ln K} - 1$), and then can be characterized by large non-linearity to ERQ. The non-linearity of $\Delta p/p_0$ is higher and the extinction probability increases more sensitively for a small increase of ERQ when the carrying capacity is larger and the environmental variance is smaller. Quasi-linearity could be achieved only when $(2/\sigma_e^2)\ln K$ is much smaller than 1, which is unrealistic for natural populations of wildlife. Therefore, it is suggested that the extinction risk of chemical substances tends to be concentrated on substances with higher ecological impact.

When ERQ takes a certain value larger than 0, the larger the carrying capacity and the smaller the environmental variance, the larger the value of$\Delta p/p_0$. When the carrying capacity is large, the background

value of the extinction probability is very small, so the extinction risk indicated by the relative value to the background value is large. Even assuming a relatively small population (K = 100) and a large environmental variance ($\sigma_e{}^2$=0.1), the ecological impact of a chemical substance with an ERQ of 0.1 is $\Delta p/p_0 = 10^4$. And it is presumed that the ecological risk is to increase the extinction probability by 4 orders of magnitude.

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Supporting Information

S-1. *The size dynamics and the energy budget in the fish*

A-TERAM uses the framework of Dynamic Energy Budget（DEB）model (Kooijman and Metz 1984; Kooijman 2010) to model individual body growth and toxicants' chronic effects on the body growth. The final output of the adverse effect of chemicals is measured in terms of reduction of reproductive output (fecundity) when the focal individuals become mature.

In the DEB model, the total energy untaken by a whole body is assumed to be proportionate to the body surface area as follow,

$vfW^{2/3}$

in which, v is the intake rate of energy per unit body surface area, W is body (wet) weight, and f is the feeding rate (its maximum is 1) .

A portion of the total intake energy is allocated into respiration (metabolism) with the allocation rate constant κ. By the mass balance between the respiration energy consumption and the intake energy, we get

$$\kappa vfW^{2/3} = mW + g\frac{dW}{dt} \ , \qquad \text{(S1-1)}$$

in which, m and g are the costs of maintenance and body growth respectively. The body length L is associated with the body weight W as $W \equiv L^3$.

If foods are plenty, the maximum length L_{max} and the maximum weight W_{max} of adults with a long period after maturity are deduced as $W_{max} = (\kappa v/m)^3$ and $L_{max} = \kappa v/m$ from $f = 1$ and $dW/dt = 0$. Equation (S1-1) gives the solution as follows if all parameters are constant,

$$L(t) = L_{max} - (L_{max} - L_b)e^{-\gamma t} \qquad \text{(S1-2)}$$

where L_b is the initial (newborn neonate) body length and $\gamma = \frac{m}{3g}$, giving the von Bertalanffy equation.

However, this symbolic solution is not relevant for cases where environmental concentrations of chemicals can change with time and the model parameters are affected by the effect of chemicals. Equation (S1-1) is equivalent to

$\frac{dL(t)}{dt} = \frac{\kappa v}{3g} - \gamma L(t)$ and

$$\frac{dL(t)}{dt} = \gamma\{L_{max} - L(t)\} \ , \qquad \text{(S1-3)}$$

A-TERAM uses the discrete form of Equation (S1-3) as follows

$$L(t+1) = L(t) + \max[\gamma\{L_{max} - L(t)\}, 0]. \qquad \text{(S1-4)}$$

A-TERAM assumes that the effect of chemicals exclusively increases the maintenance cost m, because this is the simplest way to install the hazard function in Equation (S1-4), while it is well reflected by the reduction of adult body size which determines the reproductive output. Thus, the body size dynamics of individuals of age a at time t with the time-dependent hazard $f_6(t)$ of a chemical is indicated by the following difference equation.

$$L(t+1,a+1) = L(t,a) + \max\left[\gamma\left\{L_{max} - \frac{L(t,a)}{1-f_6(t)}\right\}, 0\right] \quad \text{(S1-5)}$$

In simulations, the body size dynamics during a year is determined with the fixed exposure regime, which is assumed to repeat every year, beforehand the community dynamics is executed in the next year. This means that the adult female fish which start reproduction at the spring season in the simulation are reflected by the effect of chemicals which have accumulated in the previous seasons.

The per-capita reproductive output is assumed to be proportionate to the cubic body length,

$$r(t,a) = \begin{cases} F_c\left(\frac{L(t,a)}{L_{max}}\right)^3 & if\ L(t,a) \geq L_\alpha \\ 0 & \text{otherwise} \end{cases}, \quad \text{(S1-6)}$$

in which F_c is the maximum per-capita daily fecundity and L_α is the body length at the first reproduction.

S-2. *The concentration-response function*

The response threshold, z, is conceptually assigned to each individual and assumed to be distributed among individuals in a population. The distribution of response thresholds, $p(z)$, was assumed to be the quadratic function as follows,

$$p(z) = \begin{cases} \frac{3}{2d}\left(1 - \left(\frac{z-\bar{z}}{d/2}\right)^2\right) & \text{if} \quad |z-\bar{z}| \leq \frac{d}{2} \\ 0 & \text{otherwise} \end{cases}, \quad \text{(S2-1)}$$

in which $\bar{z}$ was the mean threshold, and d was the range of threshold. The range of threshold represented variation of tolerance to toxicants among individuals within populations, and it was set unity. This meant that the tolerance varied by one order of magnitudes within a species. However, this assumption could be manipulated by changing d. The subscript for h, p, f, β and z denoting the kind of response is declined in this section as the functional form is common among different responses.

The concentration-response function was given by

$$f(x) = \int_{\bar{z}-d/2}^{\bar{z}+d/2} h(x)p(z)dz. \quad \text{(S2-2)}$$

Using equation (S2-1), the right-hand-side of the above equation was rewritten as the following expression (as for the fish, the environmental concentration x is replaced by the internal body concentration c):

$$f(x) = \int_{\max[x-\beta^{-1},\bar{z}-d/2]}^{\min[x,\bar{z}+d/2]} h(x-z)p(z)dz + \int_{\bar{z}-d/2}^{\max[x-\beta^{-1},\bar{z}-d/2]} p(z)dz, \quad \text{(S2-3)}$$

in which

$\int p(z)dz = \frac{3}{2d}\left\{z - \frac{d}{6}\left(\frac{z-\bar{z}}{d/2}\right)^3\right\}$ and $\int zp(z)dz = \frac{3}{2d}\left[\left\{\frac{1}{2} - 2\left(\frac{\bar{z}}{d}\right)^2\right\}z^2 + \frac{8\bar{z}}{3d^2}z^3 - \frac{z^4}{d^2}\right]$.

Then the right-hand-side of equation (S2-3) was further specified as

$$f(x) = \begin{cases} 1 & \text{if } x \geq \bar{z} + d/2 + \beta^{-1} \\ 0 & \text{if } x \leq \bar{z} - d/2 \\ \beta x[\mathrm{Int}F]_{\max[x-\beta^{-1},\bar{z}-d/2]}^{\min[x,\bar{z}+d/2]} - \beta[\mathrm{Int}ZF]_{\max[x-\beta^{-1},\bar{z}-d/2]}^{\min[x,\bar{z}+d/2]} + [\mathrm{Int}F]_{\bar{z}-d/2}^{\max[x-\beta^{-1},\bar{z}-d/2]} & \text{otherwise} \end{cases} \quad \text{(S2-4)}$$

in which $\mathrm{IntF} = \frac{3}{2d}\left\{z - \frac{d}{6}\left(\frac{z-\bar{z}}{d/2}\right)^3\right\}$, $\mathrm{IntZF} = \frac{3}{2d}\left[\left\{\frac{1}{2} - 2\left(\frac{\bar{z}}{d}\right)^2\right\}z^2 + \frac{8\bar{z}}{3d^2}z^3 - \frac{z^4}{d^2}\right]$,

and $[function]_{\beta}^{\alpha}$ indicated $function(\alpha) - function(\beta)$. For simulations, response rates were numerically evaluated using equation (S2-4).

S-3. *The default value of the range (d) in the response function*

Mano and Tanaka (2017; unpublished data) estimated tolerance values of 97 isofemale strains of Daphnia galeata against two insecticides (Fenvalerate and Fenitrothion) from resting eggs collected at several sampling sites in the vicinity of Lake Kasumigaura (Ibaraki, Japan). The results indicated that the mean and the standard deviation of tolerance to Fenvalerate was respectively 2.44 (log[ppt]) and 0.178 (log[ppt]), with the range as 0.846 (log[ppt]). As for Fenitrothion, the mean and the standard deviation of tolerance was respectively 2.80 (log[ppt]) and 0.285(log[ppt]), with the range as 1.326. Thus, the range of tolerance in the logarithmic scale was about 1, which provided an empirical ground of assuming the threshold in the hazard function varied by approximately 10 folds within a species.

Reference

Mano, H. and Tanaka, Y. 2017. Spatial difference in genetic variation for fenitrothion tolerance between local populations of *Daphnia galeata* in Lake Kasumigaura, Japan. Ecotoxicology 26 (10) : 1358-1365.

S-4. *The calculation procedure for the elimination constant*

The elimination constant tends to be smaller with a larger value of K_{ow} across chemicals. Hawker and Connell (1985) indicated that the elimination constant in fish (guppies, carps and trout) for hydrophobic organic chemicals that had K_{ow} values ranging from 2.6 to 6.2 could be approximated by the following equation (c.f. Barber 2003; Kooijman et al 2004),

$\log(k_e) = -0.663\log(K_{ow}) + 0.947.$ (S4-1)

We adopted two valid digits in A-TERAM, resulting in the indirect estimate of elimination constants as

$k_e \approx 10^{-0.66\log(K_{ow})+0.95}$. (S4-2)

For the case where the depuration test data was available to derive the half-life $T_{1/2}$, the elimination constant was estimated as $k_e = 1 - 10^{\frac{-\log(2)}{T_{1/2}}}$.

S-5. *The conjectured ecological parameters*

i) *Population growth rate of algae*, R_a: according to Andersen (1997), green algae of *Senedesmus* spp. and *Selenastrum* spp. has an intrinsic rate of population increase of 1.64 on mean across species with a mode of 1.68. Thus, the population growth rate is approximately 5 ($\simeq e^{1.6}$) under optimal conditions. In the model, we adopted a slightly modest value of 4 because A-TERAM postulates spring proliferation of algae when the water temperature is suboptimal.

ii) *Carrying capacity of algae*, K_a: the chlorophyll peak density in a natural lake where *Anabaena* sp. dominated was reported to be 194 μg Chla/L (Horne and Goldman, 1994). The typical phytoplankton carrying capacity was here regarded as 20 μg Chla/L under the assumption that the algal bloom of blue-green algae was one order of magnitude higher in terms of chlorophyll density than the carrying capacity of green algae, which were edible for *Daphnia* and likely to occupy a small fraction of the algal bloom (Horne and Goldman, 1994).

iii) *Maximum grazing rate of Daphnia*, G_{max}: the maximum ingestion rate of *Daphnia galeata* is approximately 1.25 μg C/individual/hour (C = carbon) (Urabe and Watanabe, 1991). If the maximum grazing rate per day corresponds to 5 times the maximum ingestion rate per hour (because the maximum ingestion rate is unlikely to be kept constant for 24 hours) and the dry body weight of *D. galeata* is approximately 0.13 mg (Kreutzer and Lampert, 1999), then the maximum grazing rate of *D. galeata* would be 48 μg C/mg Z/day (Z = zooplankton dry biomass). By applying the conversion factor for chlorophyll content to cell carbon in phytoplankton, which is 1:70 (Reynolds, 2006), an approximate value for the maximum grazing rate as measured by chlorophyll content is 0.7 μg Chla/mg Z/day. This maximum grazing rate was estimated for mature individuals at the peak of their reproductive capacity. Taking into account the age structure in populations, the model adopted a slightly smaller value 0.5 for G_{max}.

Some field observations on plankton community dynamics revealed that the maximum population density of *Daphnia* species in eutrophic lakes ranged from approximately 80 to 300 individuals per liter, which corresponded to 24 mg/L and 90 mg/L, respectively, for dry weight density (Clark and Carter 1974). The carrying capacity K_d of *Daphnia* was set 100 mg/L as a likely value of the maximum dry weight density.

iv) *Conversion coefficient from algae to Daphnia*, c: the intrinsic rate of natural increase of zooplankton, denoted here as r_z, can be achieved when the food resource is unlimited, the temperature is optimal, the density-dependent effect is negligible, and chemical toxicants do not exist. Equation (3) indicates that r_z is associated with some model parameters: $r_z = cG_{max}$, if the daily mortality is disregarded. The conversion coefficient was indirectly estimated as 0.5 because the intrinsic rate of natural increase of *Daphnia* species is known to be about 0.25 (Andersen, 1997) and the maximum grazing rate, G_{max}, was set as 0.5.

v) *Half-satiation constant for Daphnia grazing*, h_a: the threshold food concentration of *D. galeata*, which is

defined as the lowest concentration of algae that can support non-zero population growth of *Daphnia*, has been estimated to be 50 μg C/L (Lampert, 1994; Kreutzer and Lampert, 1999). If the density of algae at which *Daphnia* grazing is half-satiated is taken to be 3 times higher than the threshold food concentration, the half-satiation constant is approximately 150 μg C/L. Thus, h_a was set as 2 μg Chla/L.

vi) *Maximum per-capita daily fecundity of fish*, F_c: an inbred strain of *O. lapites* (h-drR) laid about 150 eggs per female over 14 days, whereas hybrid individuals of two inbred lines (h-drR and drR) laid about 300 eggs per female over 14 days (personal observations). Thus, a female of *O. latipes* lays 10–20 eggs per day on average. Taking the median value, the maximum daily fecundity of fish was assumed to be 15 eggs per day per individual.

vii) *Daily survival rate of fish*, $S_f(a)$: the age-specific survival rate is one of the most important parameters that decides the reference value of the annual population growth rate of fish. The daily survival rate was separately set for 2 age periods, the adult period and the immature period (eggs, larva, and juveniles), such that the simulated population effectively tracked the observed pattern of annual and seasonal dynamics for *O. latipes*. According to field surveys on wild populations inhabiting paddy field drains, the mean rate of increase in numerical abundance from adult individuals during the breeding season in early May to juvenile individuals in late July or early August was 6.5-fold. For modeling, the maximum population growth rate during this period was assumed to be 10-fold. The annual rate of increase in the number of individuals that we observed in May in successive years was 1.7 on average (with a median value of 0.46); the maximum value was about 3 when a few extreme values were disregarded. Thus, the offspring population in mid-summer is more than 5 times more abundant on average than the reproducing parent population, and it decreases by more than 2 thirds for 9 months afterwards until the next breeding season. From this, it can be inferred that ${S_{f(a)}}^{270} = 1/3$ and $S_{f(a)} = 0.996$, which was set as the baseline value for the daily survival rate of mature fish ($\alpha_m \leq a \leq \alpha_{max}$). The age of maturity α_m and the lifespan α_{max} was set as 71 and 420 days, respectively, because this species starts reproduction about 2.5 months after hatching. The population growth rate from April 1st to August 1st (122th day), if all females reproduce from the 22nd day (April 22nd) for 50 days, is calculated as $F_c \sum_{t=22}^{71} \left[S_{f(mature)}^{\max(t-1,51)} S_{f(juvenile)}^{\min(122-t-1,70)} \right]$, in which $S_{f(juvenile)}$ and $S_{f(mature)}$ are the juvenile and the adult survivorship. Using $S_{f(mature)} = 0.996$, the following equation, $F_c \sum_{t=22}^{71} \left[S_{f(mature)}^{\max(t-1,51)} S_{f(juvenile)}^{\min(122-t-1,70)} \right] = 10$, specifies the daily survivorship for immature individuals $S_{f(juvenile)} = 0.94$ if the period from egg hatching to the end of juvenile period is assumed to be 70 days.

viii) *Body growth rate of fish, γ; asymptotic maximum body length of fish, L_{max}; and body length at the first reproduction, L_α*: a field survey on a natural population of *O. latipes* measured body lengths on several occasions within a year. The body length data were best fit to a growth function that was similar to equation (11): $L(t) = C_L \exp(-\gamma t) + L_{max}$, in which C_L is a coefficient (the function of $L(t)$ is defined for the range of t where $L(t)$ has a positive value, otherwise $L(t) = 0$), and gave the best estimate of γ as 0.00914. *O. latipes* are likely to reach their maximum body size, which is nearly 30 mm, in the breeding season of their second year. Using $\gamma = 0.00914$ and $t = 426$ (early June in the next year) in the growth function gives a body length prediction of 29.02 mm, which is compatible with field observations. Thus, the body growth

rate γ and the maximum body size L_{max} were set as 0.00914 and 29 mm respectively. To assume the length at birth L_b as 2 mm, the hypothetical date of birth in the field was specified as the 26th day counting from April 1st. If the age of first reproduction of this species in the field is about 120 days, a growth function specified as $L(120 + 26)$ gives an estimated body length at maturity of 20.4 mm. Therefore, the body length at first reproduction L_α was set as 20 mm in the simulation.

ix) *Feeding niche width of fish*, ω; *half-satiation constant in predation by fish*, h_d: these two parameters determine the relative importance of the direct effect on fish and the indirect effect through *Daphnia* or algae. Unfortunately, these parameters lack any empirical grounds. We arbitrarily set $\omega = 0.25$ and $h_d = 5$, because these values are plausible and gave a λ value of 2.18, which is compatible with the field observation.

S-6. *The determination of response slope of each endpoint based on the Ecotox-MoE data*

A-TERAM did not use values of the response slope η that were individually determined from each set of test data. The response slope is one of the most important parameters in the concentration-response function, since it characterizes the response shape and greatly affects the results of effect evaluation. However, estimates of η for any vital properties based on individual sets of ecotoxicity testing are likely to have large uncertainties, because even the standard toxicity tests that are consistent with the test guidelines are not necessarily designed, in terms of the number of treatments and replicates, to produce precise estimation of the response slope. Therefore, A-TERAM used the most frequent response-specific values of η for all chemicals in principle.

Both the no-observed effect concentration (NOEC) and the 50 % effect (or lethal) concentration (EC_{50} or LC_{50}) are available for so many substances from Ecotox-MoE (see Tanaka et al. 2020, Supplemental Data 3) that it is possible to determine the response slope parameter η for each endpoint by each chemical, and to infer the response-specific distribution of η, using the individual linear hazard function, $h(x) = \min[1, \eta\max[0, x - z]]$, and the population-level concentration-response function, $f(x) = \int_{\bar{z}-d/2}^{\bar{z}+d/2} h(x)p(z)dz$, where $p(z)$ is a quadratic function.

The numbers of sets of data available for estimating the response slopes are 341 the algae growth inhibition, 480 for the *Daphnia* acute immobility, 239 the *Daphnia* reproductive inhibition, 358 the fish acute mortality, and 34 for the fish growth inhibition (ELS test; Supporting Information S-7).

As for the algae growth inhibition and the *Daphnia* reproductive inhibition, the response slope was calculated as $\eta = 0.5/(\log EC_{50} - \log NOEC)$. As for the *Daphnia* acute immobility, since the response was defined with the daily unit while the test period in Ecotox-MoE was set 2 days, the slope parameter was determined from $\{1 - \eta(\log EC_{50} - \log NOEC)\}^2 = 0.5$ and then $\eta = 1 - \sqrt{0.5}/(\log EC_{50} - \log NOEC)$.

As for the fish acute mortality, accumulation of chemicals in the fish body was taken into account in deriving the concentration-response function, such that the toxicant concentration was expressed in terms of body burdens to keep the consistency with the concentration-response function used for the fish

population model. However, the default value of k_e=0.2 was used as the depuration constant, because highly bioaccumulative substances were not included in Ecotox-MoE. From equation (7b), the concentrations in the fish body under constant exposures of LC_{50} and NOEC for t days were respectively denoted in the logarithmic scale as $c_{LC50}(t) = \log[k_E LC_{50} \sum_{\tau=1}^{t}(1-k_E)^{\tau-1}]$ and $c_{NOEC}(t) = \log[k_E NOEC \sum_{\tau=1}^{t}(1-k_E)^{\tau-1}]$. When the test (or constant exposure) period is D days, the internal mean threshold was derived as $\bar{z} = c_{NOEC}(D)$. The response slope was estimated as the numerical solution of the following equation, $\prod_{\tau=1}^{D}\{1-\min[1,\beta \max(0, c_{LC50}(\tau)-\bar{z})]\} = 0.5$, since the response rate at the end of the test period was predicted as $\prod_{\tau=1}^{D}\{1-\min[1,\beta \max(0, c_{LC50}(\tau)-\bar{z})]\}$. As for the fish growth inhibition, the procedure to get β values is presented in Supplemental Data 9, as an additional reanalysis of the ELS data was required.

Estimates of response slopes were fit to the log-normal distribution with the least square method for each vital response. The density function of η used as the model here was $p(\eta) = \frac{1}{\sqrt{2\pi v}} exp\left\{-\frac{(\ln\eta - m)^2}{2v}\right\} d\eta$, in which m is the mean and v is the variance. The observed frequencies of η values fallen into 10 or 30 classes (the number of classes depended on the sample size and the range) were best fit to the predicted densities, to estimate the best estimate of these parameters. The mode values are equal to $exp(m\text{-}v)$. The best-fit estimates of these parameters for each response are the following:

m = - 0.478, v = 0.177, mode = 0.519 for the algae growth inhibition (sample size: 341),

m = - 0.11, v = 0.272, mode = 0.682 for the *Daphnia* acute immobility (sample size: 480),

m = 0.11, v = 0.276, mode = 0.847 for the *Daphnia* reproductive inhibition (sample size: 239),

m = 0.248, v = 0.739, mode = 0.612 for the fish acute mortality (sample size: 358), and

m = - 0.843, v = 0.149, mode = 0.371 for the fish growth inhibition (sample size: 34).

The graphical representations of the estimated response slopes plotted against the log-normal distribution are given below and in the main text (Fig. 2-7).

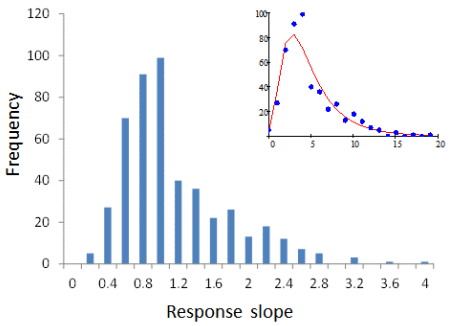


Figure S6-1. The histogram and the best-fit log-normal distribution for the response slope η in the *Daphnia* immobility. The concentration-response data were based on Ecotox-MoE.

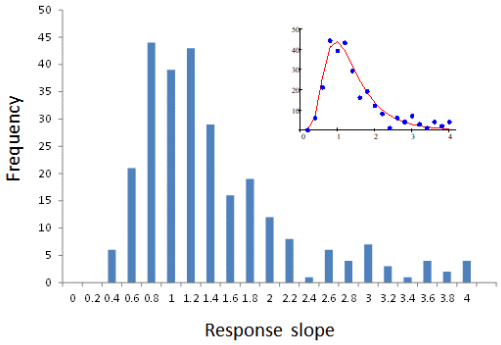


Figure S6-2. The histogram and the best-fit log-normal distribution for the response slope η in the *Daphnia* reproductive inhibition. The concentration-response data were based on Ecotox-MoE.

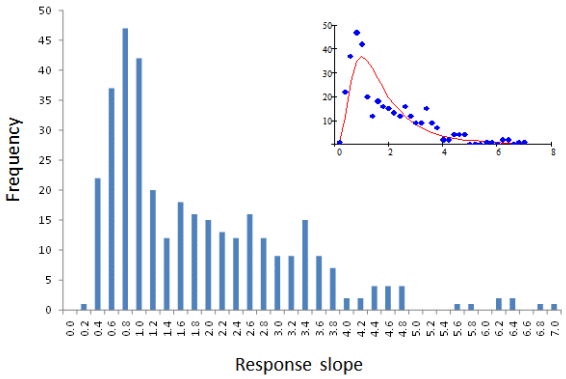


Figure S6-3. The histogram and the best-fit log-normal distribution for the response slope η in the fish acute mortality. The concentration-response data were based on Ecotox-MoE.

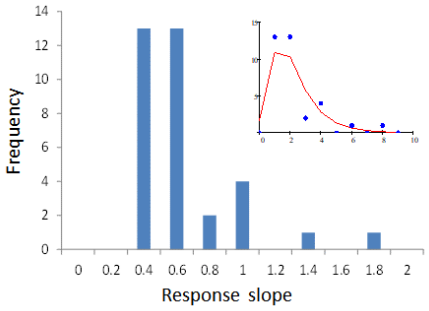


Figure S6-4. The histogram and the best-fit log-normal distribution for the response slope η in the fish growth inhibition. The concentration-response data were based on Ecotox-MoE.

Reference

Barnthouse, L. W. and G. W. Suter II. 1986. User's Manual for Ecological Risk Assessment. ORNL-6251. Oak Ridge National Laboratory.

Tanaka, Y., S. Oda., K. Nakamura and N. Suzuki 2020. A 3-species aquatic community model for ecological risk assessment using basic ecotoxicity data. Environmental Toxicology and Chemistry 29: 1086-1100.

S-7. *The procedure to estimate response slope of growth inhibition in fish from the fish early life stage data (ELS)*

Here I describe the procedure I used to obtain information on the occurrence distribution of response slopes of fish growth inhibition based on the fish early-stage test data included in Ecotox-MoE.

The fish growth is assumed to subject to von Bertalanffy function. The response slope in terms of body growth of fish is determined with the growth model because the hazard function is implemented in the model parameters of the growth model. In addition, since the exposure period is generally long in the ELS test, the model must include bioaccumulation in the fish body. We retrieved the required data of BCF and K_{ow} for characterizing the bioaccumulation process from other databases including EU IUCLID (International Uniform Chemical Information Data Base) Data Sheet; J-CHECK in National Institute of Technology and Evaluation, Incorporated Administrative Agency, because Ecotox-MoE does not contain such data. Because k_e was not available from those databases, it was indirectly estimated from K_{ow} in the case where BCF was equal to or larger than 100.

In ELS test, the exposure period is divided into the period before hatching of larva (egg stage) and the period of larva after hatching. Depending on the test, the exposure of pre-hatching period was often not practiced. In A-TEAM, the bioaccumulation process rate is common between the pre-hatch and post-hatch periods. And the dilution of chemicals by body growth was assumed negligible. Denote the pre-hatch

period in day is denoted as D_{pre} and the post-hatch period as D_{post}. The total exposure period is then D_{pre} + D_{post}. The fish body length is measured at the end of the exposure period. The body length measurement is conducted also for the fish of control at the same time as the fish under treatment.

I used only NOEC of body length reduction as the toxicity endpoint in the ELS test. The mean response threshold $\bar{z}$ and the internal concentration of chemicals $c_x(t)$ by constant exposure of concentration of x until t-th day after hatching were represented as

$$\bar{z} = \log\left[k_E \sum_{\tau=0}^{D_{pre}+D_{post}-1}(1-k_E)^{\tau} NOEC\right] \quad \text{(S7-1)}$$

$$c_x(t) = \log\left[k_E \sum_{\tau=0}^{D_{pre}+t-1}(1-k_E)^{\tau} x\right]. \quad \text{(S7-2)}$$

Estimation of the response slope in the hazard function requires an estimate of exposure concentration (EC_x) that causes a certain reduction of body length. Then, I derived concentrations of chemicals (EC_{20}), based on the ELS tests of Ecotox-MoE, that caused 20 percent reduction of body length in comparison to the control at the end of test (the data were provided by the Environmental Risk Assessment Office, Environmental Health Department, Ministry of the Environment).

EC_{20} values were estimated using linear regression equations of fish (medaka) body length on exposure concentrations for 34 chemical substances in the ELS tests (see T-1). As for 1-chloro-octane, fitting to the linear regression was not successful then a non-linear regression with a quadratic equation was applied to derive EC_{20}.

The fish body length in the control varied between different tests although the test condition and species were nearly completely unified, which was likely reflected by strong sensitivity of growth rate of the test species (*Oryzias latipes*) depending on uncontrollable trivial test condition. To exclude the effect of differences in the baseline growth rate between tests to the estimated toxicant effect to the body growth rate and to the response slope in terms of body growth rate, I estimated the body growth rate in the control condition without exposure to chemicals and determined the net reduction of growth rate due to exposure as the difference from the baseline growth rate in each data set. The body length at the start of test and the maximum body length was respectively assumed as L_0 = 2 (mm) and L_{max} = 29 (mm). The body length in the control at the end of test L_c is subject to the following equation,

$L_c = L_{max} - (L_{max} - L_0)e^{-\gamma D_{post}}$.

Using the measure body length in the control, numerical calculation (with the iterative method using 0.009 as the seed of γ) estimated the body length in the control L_c for each test.

The body length $L_x(t)$ at the t-th day after hatch under the stationary exposure of concentration x is written below according to the hazard function under the same exposure, $h_x(t) = \min\big(1, \eta\max(0, c_x(t) - \bar{z})\big)$:

$$L_x(t+1) = L_x(t) + \max\left[\gamma\left\{L_{max} - \frac{L_x(t)}{1-h_x(t)}\right\}, 0\right] . \quad \text{(S7-3)}$$

The above equation is rewritten as the following iterative equation, if $h_x(t) \le 1 - L(t)/L_{max}$ (Re: $L_x(0) = L_0$, $h_x(0) = 0$),

$$L_x(t+1) = \left(1 - \frac{\gamma}{1-h_x(t)}\right) L_x(t) + \gamma L_{max} \quad \text{(S7-4)}$$

Solving the iterative form gives

$$L_x(1) = (1-\gamma)L_0 + \gamma L_{max} \quad \text{(S7-5)}$$

and

$$L_x(t) = L_0 \prod_{\tau=0}^{t-1}\left(1 - \frac{\gamma}{1-h_x(\tau)}\right) + \gamma_0 L_{max} \sum_{s=1}^{t-1}\prod_{\tau=s}^{t-1}\left(1 - \frac{\gamma}{1-h_x(\tau)}\right) + \gamma L_{max}. \quad \text{(S7-6)}$$

Therefore, the expected body length $\hat{L}_{EC20}$ under the constant exposure of EC_{20} is

$$\hat{L}_{EC20} = L_0 \prod_{\tau=0}^{D_{post}-1}\left(1 - \frac{\gamma}{1-h_{EC20}(\tau)}\right) + \gamma L_{max} \sum_{s=1}^{D_{post}-1}\prod_{\tau=s}^{D_{post}-1}\left(1 - \frac{\gamma}{1-h_{EC20}(\tau)}\right) + \gamma L_{max}. \quad \text{(S7-7)}$$

Here, $h_{EC20}(t)$ is the hazard function when the constant exposure concentration is equivalent to EC_{20}. I determined the response slope of the hazard function such that it returned the predicted response that was compatible with EC_{20} in terms of fish body length.

S-8. *The compound effect of multiple chemicals*

For the case where compound effect by multiple chemicals is concerned, the compound effect model which are based on no interaction effect can be implemented into the risk assessment by A-TERAM. The two alternative basic compound effect models, the independent action model and the concentration addition model, are used as the module for mixture effect in A-TERAM.

(i) Independent action model

Each chemical independently affects the individual-level hazard. The total hazard function $\bar{H}(\mathbf{x})$ including entire component chemicals in the mixture is described as follows:

$$\bar{H}(\mathbf{x}) = 1 - \prod_{i=1}^{N_c}\left(1 - \int_{\bar{z}_i - d/2}^{\bar{z}_i + d/2} h_i(x_i) f_i(z) dz\right), \quad \text{(S8-1)}$$

and

$$\bar{H}(\mathbf{x}) = 1 - \prod_{i=1}^{N_c}\left(1 - \bar{h}(x_i|\bar{z}_i, \eta_i)\right). \quad \text{(S8-2)}$$

Here the subscript i denotes i-th component, N_c the total number of component and $\mathbf{x}$ time-series data of exposure concentrations. Numerical calculations in the model use equation (S8-2). The hazard functions for fish which process internal concentrations of chemicals follow the same procedure to make the total hazard function of the mixture.

(ii) Concentration addition model

For the case where the concept of concentration addition holds and the simulation is deterministic, the mixture effect is predicted by the addition of concentration of each component scaled by the threshold concentration, because the response slope is set the mode value and is identical across components. Then, the hazard function is

$$H_m(\mathbf{x}) = \min\left[1, \eta \max\left(0, \log \sum_{i=1}^{N_c} X_i^*\right)\right], \quad \text{(S8-3)}$$

where $X_i^* = \frac{X_i}{\theta_i}$ and X_i is the exposure concentration of the i-th component in the non-transformed scale θ_i is the response threshold of the i-th component (without being transformed into the logarithmic scale), which is assumed to be NOEC values except for fish and equivalent to the response threshold if logarithmically transformed, $\bar{z}_i = \log \theta_i$.

For the case of stochastic simulation, each component chemical is assigned a response slope which is randomly sampled from the log-normal distribution, and then each chemical is assumed to have different response slope. For such case the weighted average of response slopes among all components can be used to predict mixture effect according to the generalized concentration addition approach (Tanaka and Tada 2018). The weighting is $w_i = -\frac{X_i}{\theta_i} \ln\left(\frac{X_i/\theta_i}{\sum_{i=1}^{N}(X_i/\theta_i)}\right)$, then the adjusted response slope by the mixture effect is

$$\eta_m = \frac{\sum_{i=1}^{N_c} w_i \eta_i}{\sum_{i=1}^{N_c} w_i}. \quad \text{(S8-4)}$$

The concentration of each component is transformed into the following metric,

$$X_i^* = \left(\frac{X_i}{\theta_i}\right)^{\eta_i/\eta_m} . \quad \text{(S8-5)}$$

Using the transformed metric, the hazard function for the mixture is subject to the following expression,

$$H_m(\mathbf{x}) = \min\left[1, \eta_m \max\left(0, \log \sum_{i=1}^{N_c} X_i^*\right)\right] . \quad \text{(S8-6)}$$

The hazard function for the entire population of test organisms is given by

$$\bar{H}_m(\mathbf{x}) = \int_{-d/2}^{d/2} H_m f(z) dz. \quad \text{(S8-7)}$$

This hazard function is parallel to all responses by three species except for fish responses that require some modification to use body burdens of chemicals rather than environmental concentrations.

S-9. *Examples of model parameterization for the seasonal variation scheme (2.3.3) based on observed patterns of seasonally changing environmental concentrations of chemicals*

The 4 parameters of equation (29), X_{max}, T_p, σ and k were estimated as default values based on the environmental concentrations of 8 agrochemicals (5 herbicides and 3 insecticides) monitored at Kokai river (Ibaraki, Japan) on 1991 (Special Research Report of National Institute for Environmental Studies, SR-19-95; N. Hatakeyama personal communications). The parameters were determined with the least square method. In addition to the model parameters, a measure of discrepancy from the model, *sd*, was evaluated as the standard deviation of observed concentrations from the expected concentrations scaled by the maximum concentration.

Chemicals	X_{max} (mg/L)	T_p	σ	k	*sd*
Simetrin	7.22×10^{-3}	63.6	11.5	1.88	0.094
Butachlor	1.77×10^{-3}	43.2	8.2	1.6	0.079
Pretilachlor	5.77×10^{-3}	44.6	5.4	1.05	0.035

Benthiocarb	7.00×10^{-3}	62.8	8.7	1.01	0.082
Molinate	19.01×10^{-3}	62.6	9.2	1.96	0.090
Iprobenfos	3.01×10^{-3}	97.2	15.6	1.39	0.145
Pyridaphenthion	8.94×10^{-3}	67.4	6.6	1.15	0.112
Malathion	3.72×10^{-3}	61.5	10.4	1.21	0.101

The date of maximum daily concentration T_p tended to be later for insecticides than herbicides, and the variability of concentrations was larger for insecticides than herbicides. The skewness of the variability of concentrations k was only slightly larger than 1 but smaller than 2 for all chemicals, indicating the temporal distribution of those chemicals is mostly unimodal and more skewed (concentrated) than the normal distribution.

S-10. *Transformation from ERQ to extinction probability of a population*

Transforming both sides of equation (31) into the logarithmic scale, the mean extinction time is denoted in the logarithmic scale as follows,

$$\log \bar{T} \propto \frac{2E[\ln \bar{\lambda}]}{\sigma_e^2} \log K. \quad \text{(S10-1)}$$

The decrement of population growth rate due to exposure to a chemical (the exposure concentration is x), $\Delta \log_{10} \bar{T}$, may be approximated as follows if the toxicant concentration is low and then the change in population growth rate is small,

$$\Delta \log \bar{T} \cong \frac{2 \log K}{\sigma_e^2} \frac{d \ln \bar{\lambda}}{dx} x. \quad \text{(S10-2)}$$

When we assume that the reduction in the population growth rate by chemicals is proportional to the reduction in the population growth rate in the long term, the ERQ in the present framework is equivalent to $\frac{d \ln \bar{\lambda}}{dx} x$ or $\frac{d\bar{\lambda}}{\bar{\lambda} dx} x$ in the magnitude with the opposite sign. Therefore, we can get $\Delta \log \bar{T} \cong -\frac{2 \log K}{\sigma_e^2} ERQ$.

If the extinction event is independent of time, the increment of extinction probability Δp is approximated by the following equation (Tanaka 2003),

$$\Delta p \cong p_0 \left(10^{-\Delta \log \bar{T}} - 1\right), \quad \text{(S10-3)}$$

where p_0 is the background level of extinction probability.

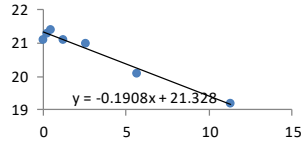
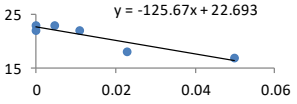
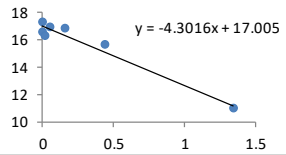
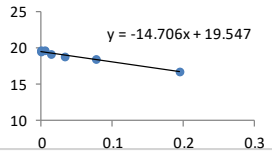
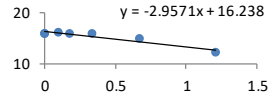
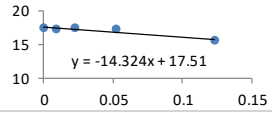
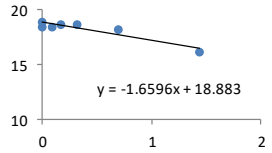
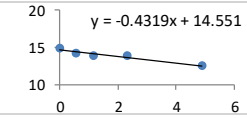
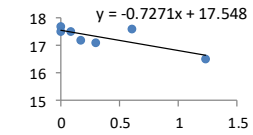
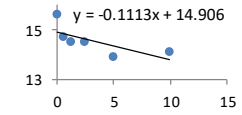
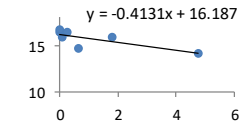
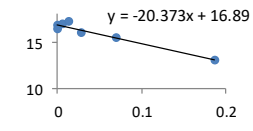
Therefore, the excessive extinction risk induced by pollutants can be related to ERQ with the following equation,

$$\Delta p \cong p_0 \left(K^{(2/\sigma_e^2) ERQ} - 1\right) . \quad \text{(S10-4)}$$

T-1 Early life stage (ELS) test data and the related statistics (from Ecotox-MoE)

CAS	Substances	KOW EXP	Log BCF EPI	Log BCF EXP	Test duration	Test duration after hatching	NOEC (mg/L)	ke	Z	EC20	η	Lc	γ
67663	Chloroform	1.97	0.697		40	30	2.61	0.2	0.417	2.23E+01	0.347	21.3	0.0418
84151	o-Terphenyl	5.52	3.31		40	30	0.011	0.002	-3.072	3.63E-02	0.73	22.7	0.0485
85687	Benzyl phthalate	4.73	2.788		42	30	0.154	0.2	-0.813	7.90E-01	0.557	17	0.027
87865	Pentachlorophenol	3.32	3.045	224	40	30	0.013	0.057	-1.930	2.65E-01	0.278	19.5	0.0348
92524	Biphenyl	4.01	2.313	280	40	30	0.338	0.02	-0.727	1.09E+00	0.972	16.2	0.0249
97007	1-chloro-2,4-dinitrobenzene	2.17	1.099	<44	38	29	0.052	0.2	-1.280	2.45E-01	0.576	17.5	0.0294
99876	p-Cymene	4.1	2.372		40	31	0.69	0.018	-0.448	2.28E+00	0.846	18.9	0.0317
104949	p-Anisidine	0.95	0.5		38	28	0.559	0.2	-0.253	6.76E+00	0.41	14.6	0.0225
106467	p-dichlorobenzene	3.44	1.937	720	40	30	0.601	0.048	-0.287	4.81E+00	0.455	17.5	0.0284
106490	p-Toluidine	1.39	0.738	<13	40	30	0.598	0.2	-0.223	2.68E+01	0.264	14.9	0.0217
108907	Chlorobenzene	2.84	1.541		43	30	0.247	0.2	-0.607	7.85E+00	0.273	16.2	0.0249
111659	Octane	5.18	3.085		41	30	0.0278	0.0034	-2.440	1.66E-01	0.6	16.9	0.0268
111853	1-chlorooctane	4.52	1.278		40	30	0.161	0.2	-0.793	2.88E-01	1.7	15.4	0.0229
117817	Di-2-ethylhexyl phthalate	7.6	3.234	29.7	40	30	0.56	0.2	-0.252	5.06E+00	0.325	22.1	0.0455
122349	Simazine	2.18	0.588		40	30	1.99	0.2	0.299	1.08E+01	0.561	16.1	0.0246
123308	4-aminophenol	0.04	0.5	3.2	41	30	0.13	0.2	-0.886	9.01E-01	0.375	21.8	0.0441
124481	Dibromochloromethane	2.16	1.092	9.2	41	30	1.05	0.2	0.021	6.84E+00	0.458	18.3	0.0309
127184	Tetrachlorethylene	3.4	1.91	77.1	39	30	1.01	0.2	0.004	6.07E+00	0.459	19.2	0.0338
128370	2,6-di-t-butyl-4-methylphenol	5.1	2.81	2800	42	32	0.0528	0.0038	-2.108	7.10E-01	0.418	15.7	0.0221
129000	Pyrene	4.88	2.887	457	39	30	0.00493	0.0054	-3.028	1.53E-02	0.909	19.7	0.0355
132650	Dibenzothiophene	4.38	2.557		40	30	0.0282	0.011	-1.996	3.99E-01	0.354	18.4	0.0312
140669	4-t-octylphenol	5.28	3.148	469	40	30	0.0334	0.0029	-2.436	3.05E-01	0.48	16.6	0.0259
141435	2-aminoethanol	-1.31	0.5		41	30	1.24	0.2	0.093	2.85E+01	0.284	17.5	0.0284
606202	2,6-dinitrotoluene	2.1	1.053	21.2	41	30	0.129	0.2	-0.889	2.91E+00	0.302	16.3	0.0251
611198	2-chlorobenzyl chloride	3.44	1.936		40	30	0.0461	0.2	-1.336	1.44E-01	0.8095	16.8	0.0265
782741	2,2'-Dichlorohydrazobenzene	4.34	2.534		41	30	0.00417	0.012	-2.788	6.87E-02	0.341	17.8	0.0293
793248	N- (1,3-dimethylbutyl) -N'-phenyl-1,4-benzenediamine	4.68	2.755		41	30	0.00371	0.0073	-3.017	3.70E-02	0.447	16.9	0.0268
1806264	p-Octylphenol	5.5	1.92		43	30	0.0077	0.2	-2.114	5.80E-02	0.34	22.9	0.0496
3380345	Triclosan	4.76	2.808		38	29	0.0603	0.2	-1.220	2.34E-01	0.632	18.4	0.0322
4170303	Crotonaldehyde	0.6	0.5	3.2	41	30	0.0247	0.2	-1.607	2.57E-01	0.38	17.5	0.0284
6165511	1, 4-dimethyl-2- (1-phenylethyl) benzene	5.39	3.127	1200	40	30	0.0338	0.0025	-2.492	3.32E-01	0.47	16.3	0.0251
7791200	Nickel chloride · hexahydrate	NA	0.5		43	30	1.05	0.2	0.021	5.68E+00	0.39	23.7	0.0543
13048334	Acrylic acid hexamethylene ester	3.08	1.698		39	29	0.0723	0.2	-1.141	1.46E-01	1.23	18.3	0.0319
25154523	Nonylphenol	5.76	2.093	330	43	30	0.0333	0.0014	-2.711	2.06E-01	0.587	16.8	0.0265

T-2 Concentration response relation in the early life stage test data (from Ecotox-MoE)

CAS	Substances		measured	TL(mm)	
67663	Chloroform	control	0	21.1	
		conc-1	0.209	21.3	
		conc-2	0.471	21.4	
		conc-3	1.23	21.1	
		conc-4	**2.61**	21	
		conc-5	5.65	20.1	
		conc-6	11.3	19.2	
84151	o-Terphenyl	control	0	23	
		solvent	0	22	
		conc-1	0.0048	23	
		conc-2	**0.011**	22	
		conc-3	0.023	18	
		conc-4	0.05	17	
85687	Benzyl phthalate	control	0	17.3	
		solvent	0	16.5	
		conc-1	0.018	16.3	
		conc-2	0.051	16.9	
		conc-3	**0.154**	16.8	
		conc-4	0.435	15.6	
		conc-5	1.35	11	
87865	Pentachlorophenol	control	0	19.5	
		solvent	0	19.7	
		conc-1	0.005	19.7	
		conc-2	**0.013**	19.2	
		conc-3	0.0323	18.8	
		conc-4	0.0762	18.5	
		conc-5	0.195	16.7	
92524	Biphenyl	control	0	15.9	
		solvent	0	15.9	
		conc-1	0.0991	16	
		conc-2	0.174	15.7	
		conc-3	**0.338**	15.8	
		conc-4	0.671	14.8	
		conc-5	1.21	12.2	
97007	1-chloro-2,4-dinitrobenzene	control	0	17.4	
		conc-1	0.00866	17.2	
		conc-2	0.0224	17.3	
		conc-3	**0.052**	17.1	
		conc-4	0.123	15.6	
99876	p-Cymene	control	0	18.5	
		solvent	0	18.9	
		conc-1	0.0865	18.5	
		conc-2	0.167	18.7	
		conc-3	0.316	18.7	
		conc-4	**0.69**	18.2	
		conc-5	1.44	16.2	
104949	p-Anisidine	control	0	14.8	
		conc-1	**0.559**	14.1	
		conc-2	1.15	13.8	
		conc-3	2.32	13.8	
		conc-4	4.9	12.4	
106467	p-Dichlorobenzene	control	0	17.7	
		solvent	0	17.5	
		conc-1	0.0867	17.5	
		conc-2	0.168	17.2	
		conc-3	0.299	17.1	
		conc-4	**0.601**	17.6	
		conc-5	1.23	16.5	
106490	p-toluidine	control	0	15.6	
		conc-1	**0.598**	14.7	
		conc-2	1.18	14.5	
		conc-3	2.47	14.5	
		conc-4	4.99	13.9	
		conc-5	9.93	14.1	
108907	Chlorobenzene	control	0	16.4	
		solvent	0	16.7	
		conc-1	0.093	15.9	
		conc-2	**0.247**	16.4	
		conc-3	0.633	14.7	
		conc-4	1.8	15.9	
		conc-5	4.76	14.2	
111659	Octane	control	0	16.8	
		solvent	0	16.5	
		conc-1	0.0057	17	
		conc-2	0.0127	17.2	
		conc-3	**0.0278**	16	
		conc-4	0.0686	15.5	
		conc-5	0.186	13.1	

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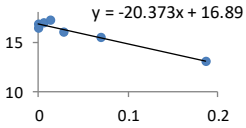
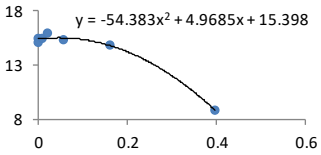
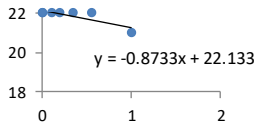
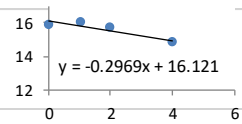
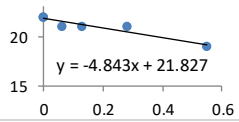
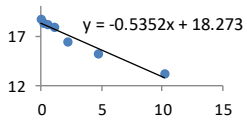
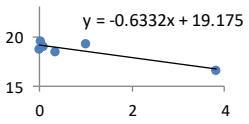
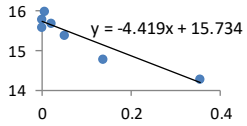
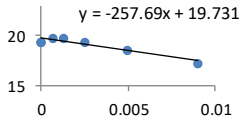
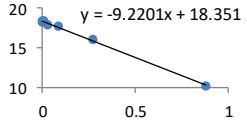
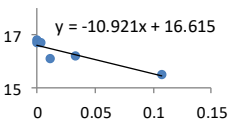
111659	Octane	control	0	16.8	
		solvent	0	16.5	
		conc-1	0.0057	17	
		conc-2	0.0127	17.2	
		conc-3	**0.0278**	16	
		conc-4	0.0686	15.5	
		conc-5	0.186	13.1	
111853	1-chlorooctane	control	0	15.4	
		solvent	0	15.1	
		conc-1	0.0085	15.5	
		conc-2	0.0206	15.9	
		conc-3	0.0567	15.3	
		conc-4	**0.161**	14.8	
		conc-5	0.397	8.8	
117817	Di-2-ethylhexyl phthalate	control	0	22	
		solvent	0	22	
		conc-1	0.11	22	
		conc-2	0.2	22	
		conc-3	0.34	22	
		conc-4	**0.56**	22	
		conc-5	1	21	
122349	Simazine	control	0	15.9	
		conc-1	1.02	16	
		conc-2	**1.99**	15.7	
		conc-3	4.01	14.8	
123308	4-aminophenol	control	0	22	
		solvent	0	22	
		conc-1	0.064	21	
		conc-2	**0.13**	21	
		conc-3	0.28	21	
		conc-4	0.55	19	
124481	Dibromochloromethane	control	0	18.7	
		conc-1	0.208	18.3	
		conc-2	0.476	18.2	
		conc-3	**1.05**	17.9	
		conc-4	2.1	16.4	
		conc-5	4.67	15.2	
		conc-6	10.2	13.2	
127184	Tetrachlorethylene	control	0	18.7	
		conc-1	0.0318	19.6	
		conc-2	0.0847	19	
		conc-3	0.351	18.5	
		conc-4	**1.01**	19.3	
		conc-5	3.81	16.6	
128370	2,6-di-t-butyl-4-methylphenol	control	0	15.6	
		solvent	0	15.8	
		conc-1	0.00817	16	
		conc-2	0.0203	15.7	
		conc-3	**0.0528**	15.4	
		conc-4	0.139	14.8	
		conc-5	0.354	14.3	
129000	Pyrene	control	0	19.4	
		solvent	0	19.3	
		conc-1	0.000642	19.8	
		conc-2	0.00129	19.7	
		conc-3	0.00247	19.4	
		conc-4	**0.00493**	18.6	
		conc-5	0.00896	17.2	
132650	Dibenzothiophene	control	0	18.3	
		solvent	0	18.2	
		conc-1	0.0087	18.4	
		conc-2	**0.0282**	17.9	
		conc-3	0.0875	17.7	
		conc-4	0.274	16	
		conc-5	0.877	10.2	
140669	4-t-octylphenol	control	0	16.8	
		solvent	0	16.7	
		conc-1	0.0034	16.7	
		conc-2	0.0112	16.1	
		conc-3	**0.0334**	16.2	
		conc-4	0.107	15.5	

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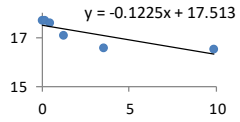
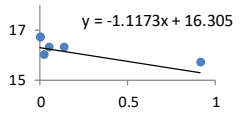
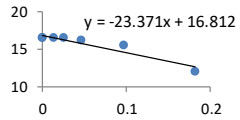
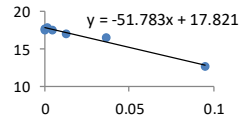
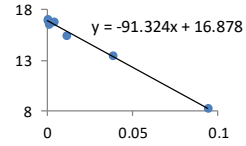
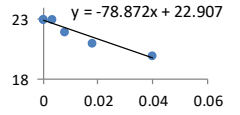
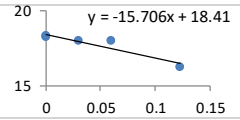
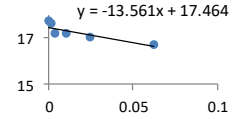
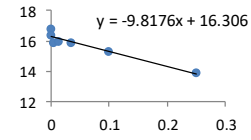
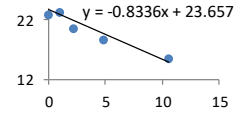
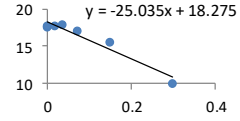
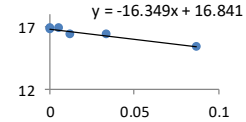
141435	2-aminoethanol	control	0	17.7	
		conc-1	0.21	17.7	
		conc-2	0.47	17.6	
		conc-3	**1.24**	17.1	
		conc-4	3.55	16.6	
		conc-5	9.85	16.5	
606202	2,6-dinitrotoluene	control	0	16.7	
		solvent	0	16.7	
		conc-1	0.019	16	
		conc-2	0.05	16.3	
		conc-3	**0.129**	16.3	
		conc-4	0.355	14.8	
		conc-5	0.913	15.7	
611198	2-chlorobenzyl chloride	control	0	16.4	
		solvent	0	16.5	
		conc-1	0.0134	16.4	
		conc-2	0.0249	16.4	
		conc-3	**0.0461**	16.1	
		conc-4	0.0967	15.4	
		conc-5	0.182	12	
782741	2,2'-Dichlorohydrazobenzene	control	0	17.7	
		solvent	0	17.6	
		conc-1	0.00157	17.9	
		conc-2	**0.00417**	17.5	
		conc-3	0.0125	17.1	
		conc-4	0.0365	16.5	
		conc-5	0.0949	12.7	
793248	N- (1,3-dimethylbutyl) -N'-phenyl-1,4-benzenediamine	control	0	16.9	
		solvent	0	17	
		conc-1	0.00142	16.6	
		conc-2	**0.00371**	16.8	
		conc-3	0.011	15.5	
		conc-4	0.0385	13.5	
		conc-5	0.0937	8.3	
1806264	p-Octylphenol	control	0	23	
		solvent	0	23	
		conc-1	0.0033	23	
		conc-2	**0.0077**	22	
		conc-3	0.018	21	
		conc-4	0.04	20	
3380345	Triclosan	control	0	18.2	
		solvent	0	18.3	
		conc-1	0.03	18	
		conc-2	**0.0603**	18	
		conc-3	0.123	16.2	
4170303	Crotonaldehyde	control	0	17.7	
		conc-1	0.0014	17.6	
		conc-2	0.0036	17.2	
		conc-3	0.0101	17.2	
		conc-4	**0.0247**	17	
		conc-5	0.0623	16.7	
6165511	1, 4-dimethyl-2- (1-phenylethyl) benzene	control	0	16.8	
		solvent	0	16.4	
		conc-1	0.0053	15.9	
		conc-2	0.014	16	
		conc-3	**0.0338**	15.9	
		conc-4	0.0997	15.3	
		conc-5	0.249	13.9	
7791200	Nickel chloride · hexahydrate	control	0	22.9	
		conc-1	0.232	24.1	
		conc-2	0.476	24.5	
		conc-3	**1.05**	23.3	
		conc-4	2.26	20.5	
		conc-5	4.81	18.7	
		conc-6	10.6	15.4	
13048334	Acrylic acid hexamethylene ester	control	0	17.5	
		solvent	0	17.7	
		conc-1	0.0179	17.8	
		conc-2	0.035	17.9	
		conc-3	**0.0723**	17	
		conc-4	0.149	15.6	
		conc-5	0.298	10.1	
25154523	Nonylphenol	control	0	16.8	
		solvent	0	16.9	
		conc-1	0.0051	16.9	
		conc-2	0.0118	16.4	
		conc-3	**0.0333**	16.4	
		conc-4	0.0873	15.4	

measured : measured concentration (mg/L), TL: body length of fish (Medaka) (mm),