

A real-time biomonitoring system to detect arsenic toxicity by valve movement in freshwater clam *Corbicula fluminea*

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Abstract Arsenic (As) is the element of greatest ecotoxicological concern in aquatic environments. Effective monitoring and diagnosis of As pollution via a biological early warning system is a great challenge for As-affected regions. The purpose of this study was to synthesize water chemistry-based bioavailability and valve daily rhythm in *Corbicula fluminea* to design a biomonitoring system for detecting waterborne As. We integrated valve daily rhythm dynamic patterns and water chemistry-based Hill dose-response model to build into a programmatic mechanism of inductance-based valvometry technique for providing a rapid and cost-effective dynamic detection system. A LabVIEW graphic control program in a personal computer was employed to demonstrate completely the functional presentation of the present dynamic system. We verified the simulated dissolved As concentrations based on the valve daily rhythm behavior with published experimental data. Generally, the performance of this proposed biomonitoring system demonstrates fairly good applicability to detect waterborne As concentrations when the field As concentrations are less than 1 mg L^{-1} . We also revealed that the detection times were dependent on As exposure concentrations. This biomonitoring system could particularly provide real-time transmitted information on the waterborne As activity under various aquatic environments. This parsimonious *C. fluminea* valve rhythm behavior-based real-time biomonitoring system presents a valuable effort

to promote the automated biomonitoring and offers early warnings on potential ecotoxicological risks in regions with elevated As exposure concentrations.

Keywords Arsenic · Freshwater clam · *Corbicula fluminea* · Valve rhythm behavior · Biomonitoring · Ecotoxicology

Introduction

Nowadays, biomonitoring technologies hold exceptional promise for continuous (real-time) data collection and simultaneous measurement of ecologically relevant metal pollution in aquatic ecosystems (Hansen 2008; Zhou et al. 2008). The typical method for biomonitoring is based on living bioindicators. Many aquatic organisms such as algae, invertebrates, mussel, and fish are common used bioindicators since their sensitive, the advantage that changes in their behaviors, e.g., swimming and avoidance response pattern can be measured immediately as responses to the occurrence of contaminants. Behavior could be used as physiological activity parameters for providing ecological relevance to standard toxicity testing, that is exceptionally suitable used in the online biomonitors (van der Schalie et al. 2001; Gerhardt et al. 2005, 2006).

Bivalves are commonly preferred for biomonitoring in aquatic ecosystems because of their wide distribution, abundant, and tolerant to various environmental conditions (Zhou et al. 2008). The freshwater clam *Corbicula fluminea* are influenced by chemical contaminants on valve closure, filtration rate or burrowing into sediment. The freshwater clam reduces the feeding activity by valve closure to avoid any toxic effects of contaminants (Kadar et al. 2001). *C. fluminea* as an in situ monitoring test

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organism that valve activity has promised as biological response since it is relatively easy and inexpensive to monitor and mirror responses at environmentally realistic concentrations (Cherry and Soucek 2006; Newton and Cope 2007).

Geogenic Arsenic (As) contamination of groundwater is a major ecological and human health problem in southwestern and northeastern coastal areas of Taiwan (Liu et al. 2007). The major freshwater clam *C. fluminea* farming sites are situated at southwestern coastal areas of Taiwan. Williams et al. (2006) indicated that As could accumulate in tissues of freshwater organism, and human who consume these As contaminated tissues may result in health risk such as renal diseases. *C. fluminea* has a high market value and commercially important to Taiwan's aquaculture. Therefore, the elevated waterborne As concentration may result in the market prices reduction and the closure of clam farms.

Based on the toxicological principles in aquatic environment, the chemical bioavailability depends on the external elements such as pH, hardness, specific ions, and chemical reaction and toxicity influencing the mechanism of the bioavailability to aquaculture species. The influences and complexities of the environment are a key problem and also have to be taken into consideration, especially, the influence of organic functional groups and oxidation state of arsenicals (Shaw et al. 2007). Little is known, however, about the relationship between clam valve daily rhythm response and As bioavailability related to an As biological early warning system design. In the previous studies (Liao et al. 2008, 2009; Chen et al. 2010), we have shown that the time-dependent median effect concentration ($EC50(t)$) and valve opening behavior to As at any response time could be well predicted in general. Therefore, the environmentally relevant water chemistries could be improved to describe rigorously the bioavailability of As causing toxicity to valve opening behavior in *C. fluminea*.

As contamination is the biggest health threat in the world (Reich 2011). Effective monitoring and diagnosis of As pollution via a biological early warning system is a great challenge for As-affected regions. The purpose of this study was to synthesize water chemistry-based As activity and valve daily rhythm in *C. fluminea* to design a bio-monitoring system for detecting waterborne As. Clam valve daily rhythm dynamic pattern and water chemistry-based Hill dose–response model were integrated to build into a valvometry technique programmatic mechanism for providing a rapid and cost-effective dynamic detection system. This study employed a LabVIEW graphic control program in a personal computer to simulate and demonstrate the functional presentations of the proposed clam valve-based biomonitoring system. We hope that this study can hold high promise for practical applications in online biomonitoring for As-affected regions.

Materials and methods

Principle of system design

Previous studies have characterized the valve daily rhythm in *C. fluminea* in response to As concentration and developed a mechanistic model to describe the interaction between As activity and valve daily rhythm incorporated with water chemistry-based Hill dose–response model (Liao et al. 2008; Chen et al. 2010). This study applied these models (referring to as the As activity—*Corbicula* model) to design the real-time valve behavior biomonitoring system for detecting As toxicity.

Generally, at equilibrium thermodynamics, arsenate (As(V)) dominants in oxidative environments of most surface waters (Ferguson and Gavis 1972). Thus, As(V) being the most prevalent in most surface waters. In this study, we constructed an approach that is used to process the As activity—*Corbicula* model incorporated with the *C. fluminea* daily dynamic As(V) detection mechanism. Figure 1a illustrates the built-in database of computational algorithm. Table 1 lists the available and the essential mathematical equations used to describe the As activity—*Corbicula* model.

The descriptions of database built-in process are as following. First, the valve daily rhythm dynamic pattern in response to As(V) should be recorded to obtain the water chemistry-based time-varying As(V) concentration response profile. The valve closure response profile could be expressed by a Hill-based dose–response model for receiving the time-dependent median response As(V) concentration ($EC50(t_R)$), the time-dependent maximum response ($R_{max}(t_R)$), and time-dependent Hill coefficient ($n(t_R)$). Second, this study considered the effective As(V) concentration

Table 1 Mathematical expressions for describing As(V) activity—*Corbicula* model, clam daily closing rhythm exposed to uncontaminated and waterborne As(V) conditions (see text for the symbol descriptions)

Mathematical expressions

Time-dependent valve closure response based on As(V) activity

$$R(t_R, \{As(V)\}) = \frac{R_{max}(t_R)}{1 + \left(\frac{EC50_{As(V)}(t_R)}{[As(V)]}\right)^{n(t_R)}} \quad (T1)$$

Time-dependent Hill coefficient function in valve closure response

$$n(t_R) = 1.65 + 2.5 \exp(-t_R/0.31), \quad r^2 = 0.99 \quad (T2)$$

Time-dependent maximum response

$$R_{max}(t_R) = 1.00 - 1.06 \exp(-t_R/0.36), \quad r^2 = 0.98 \quad (T3)$$

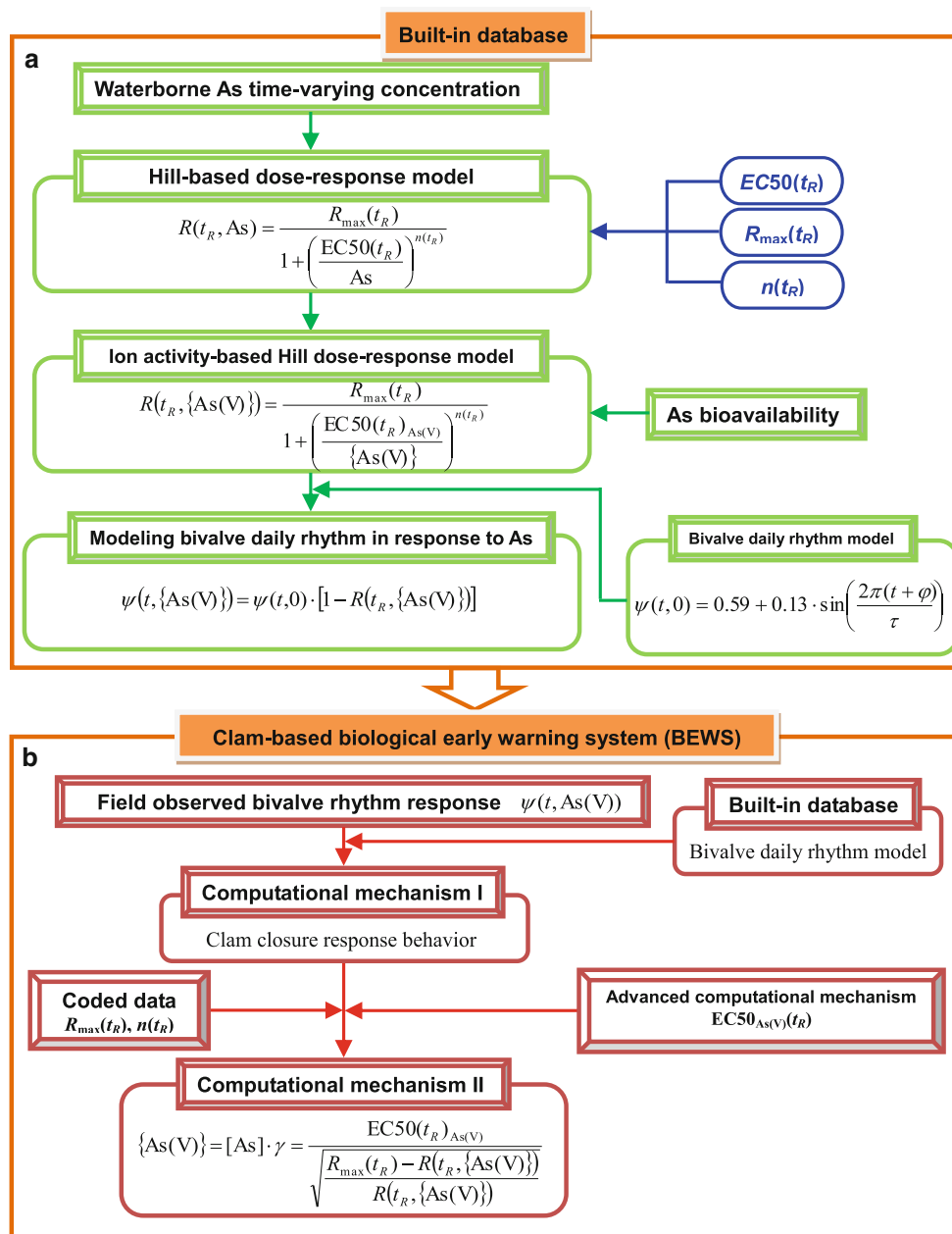
Valve daily rhythm model that with As exposure

$$\psi(t, \{As(V)\}) = \psi(t, 0) \times [1 - R(t_R, \{As(V)\})] \quad (T4)$$

Bivalve daily rhythm model that without As exposure

$$\psi(t, 0) = 0.59 + 0.13 \cdot \sin\left(\frac{2\pi(t+\phi)}{\tau}\right) \quad (T5)$$

Fig. 1 Schematic showing the framework of detection algorithm for the proposed *C. fluminea*-based biomonitoring. **a** built-in database. **b** computational algorithm of clam-based biological early warning system for As detection. (See text for detailed descriptions of symbol.)



for affecting the valve closure behavior of *C. fluminea*. The fitted functions of $R_{max}(t_R)$ and $n(t_R)$ were adopted from Liao et al. (2008). The relationship between $As(V)$ activity and $As(V)$ concentration was depended on the water chemistry. Hence the site-specific water condition could determine the $As(V)$ toxicity induced-physiological behavior for *C. fluminea*. Then, we incorporated the reconstructed the ion activities-based Hill dose–response model into the bivalve daily rhythm model that without As exposure to model the bivalve daily rhythm in response to $As(V)$ activity (Liao et al. 2009).

In the preliminary study before constructing the proposed biomonitoring system, we showed that the water ionic

concentration (e.g., calcium, magnesium, sodium, potassium, etc.) were not significantly affect the As activity based on the calculation from Windermere humic aqueous model (WHAM). However, temperature and pH were the significant factors in affecting As activity. Therefore, the measured temperature and pH information were logged into the monitoring interface of a visual window to calculate $As(V)$ activity coefficient ($\gamma_{As(V)}$). In this study, two pH range values of 6.5–7.5 and 7.5–8.2, respectively, were used to delineate the $As(V)$ activity coefficient with a temperature ranging from 15 to 40°C. The relationship among $\gamma_{As(V)}$, temperature, and pH value can be constructed by WHAM as a $\gamma_{As(V)}(temp, pH)$ response surface. Finally, we used a three-parameter liner

regression model to best fit the $\gamma_{As(V)}(temp, pH)$ response surface to obtain the fitted equations.

The valve opening dynamic mechanism $\psi(t, \{As(V)\})$ (Table 1, Eq. T4) with the variation of valve daily rhythm under various water temperature- and pH-specific As(V) activities ($\{As(V)\}$) can then be constructed. This can be achieved by integrating the bivalve daily rhythm model that without As(V) exposure $\psi(t, 0)$ (Table 1, Eq. T5) and the As activity-based dynamic bivalve responsive mechanism. Liao et al. (2009) proposed the four-parameter sine function in Eq. T5 to best describe the daily activity of valve opening in *C. fluminea* in the absence of As in that φ and τ are the phase and the daily period, respectively.

Clam-based biological early warning system

In the field situation, we need to set up the valvometry conversion technique together with temperature and pH meters to monitor bivalve rhythm response and real water conditions. The first computational mechanism used to describe the valve closure behavior in response to As(V) activity can be derived from Eq. T4 (Table 1) as,

$$R(t_R, \{As(V)\}) = \frac{\psi(t, \{As(V)\}) - \psi(t, 0)}{\psi(t, 0)}, \quad (1)$$

where $\psi(t, \{As(V)\})$ is the monitored dynamic variation of valve rhythm opening behavior exposed to As(V) activity $\{As(V)\}$ and $\psi(t, 0)$ is used as the built-in database from Liao et al. (2009).

The monitored time-varying valve closure rhythm could be used as the real As(V) activity in the field. Hence, the time-varying As(V) activity could be derived from dose-response relationship $R(t_R, \{As(V)\})$. The response time (t_R) dependent-As(V) activity can then be displayed as,

$$\{As(V)\}(t_R) = \frac{EC50_{As(V)}(t_R)}{n(t_R) \sqrt{\frac{R_{max}(t_R) - R(t_R, \{As(V)\})}{R(t_R, \{As(V)\})}}} \quad (2)$$

in that the coded data in built-in program such as $n(t_R)$ and $R(t_R)$ were be incorporated into Eq. 2. Furthermore, this study used the fitted exponential function for $EC50_{As(V)}(t_R)$ of valve closure response. Figure 1b illustrates the computational algorithm of clam-based biological early warning system.

The proposed two computational mechanisms I and II (Fig. 1b) could then be used to monitor the time-varying valve daily rhythm behavior in response to As(V) activity in the various aquatic environments.

Statistical analyses and simulation scheme

WHAM Version 6 (WHAM VI, Centre for Ecology & Hydrology, Lancaster, UK) was performed to calculate the

activities of the competing cations considered in this study. TableCurve 3D (Version 4.0, AISN Software Inc., Mapleton, OR, USA) was used to perform all statistics. Our system design processes were developed from the LabVIEW graphic control program language (Version 7.0, NI Inc., North Mopac Expressway, Austin, USA) in personal computer to monitor *C. fluminea* daily rhythm for real time detecting waterborne As(V) activity.

Results

Interface window of clam monitoring

This study performed systematically the LabVIEW-based clam monitoring interface window to measure the site-specific clam daily rhythm behavior in response to waterborne As(V). Some built-in database in program should be construed, including As(V) activity coefficient and time-dependent function of $EC50(t_R)$. The fitted three-parameter linear regression models for As(V) activity coefficient in pH values ranging from 6.5 to 7.5 and 7.5 to 8.2, respectively, have the forms as $\gamma_{As(V)}(temp, pH) =$

$$\begin{cases} \gamma_{As(V), 1}(temp, pH), & 6.5 \leq pH \leq 7.5 \\ \gamma_{As(V), 2}(temp, pH), & 7.5 < pH \leq 8.2 \end{cases}$$

$$\gamma_{As(V), 1}(temp, pH) = 8.521 \times 10^{-5} \cdot temp - 0.477 \cdot pH + 3.626, \quad r^2 = 0.98, \quad (3)$$

$$\gamma_{As(V), 2}(temp, pH) = 2.190 \times 10^{-5} \cdot temp - 0.091 \cdot pH + 0.755, \quad r^2 = 0.97, \quad (4)$$

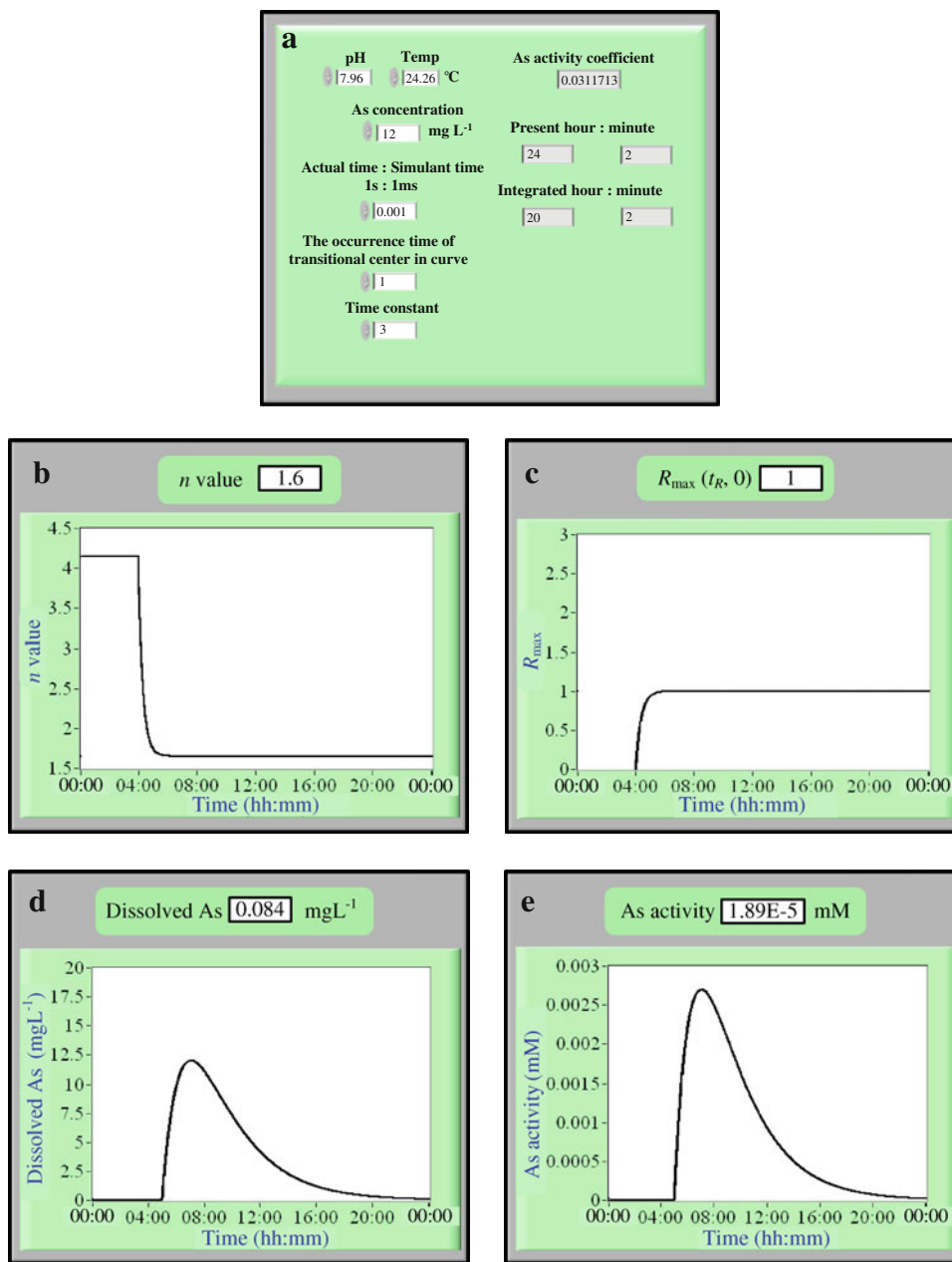
where the unit of temperature is °C. Furthermore, the fitted functions of $EC50(t_R)$ was based on valve closure behavior data from Liao et al. (2008). The exponential function of $EC50(t_R)$ can be obtained by best fitting the time-dependent EC50 values,

$$EC50(t_R) = 0.387 + 8.624 \cdot \exp(-t_R/0.239), \quad r^2 = 0.99, \quad (5)$$

where the unit of time is hour and EC50 is $mg L^{-1}$.

Figure 2a shows the interface of monitored information including pH, temperature, As(V) activity coefficient, and the forecasted waterborne As concentration in the field situation based on the valve rhythm behavior. On the other hand, the basic display information includes the proportion of actual and simulation times, the occurrence time of transitional center in curve, time constant, and the present and integrated response times. Figure 2b, c demonstrates the interfaces of built-in database $n(t_R)$ and $R_{max}(t_R)$ based on Eqs. T2 and T3 (Table 1). Figure 2d, e shows the interface of site-specific time-dependent dissolved As(V) and As(V) activity profiles, respectively. The relationship

Fig. 2 Monitoring interface showing the simulated time course of waterborne As(V) in filed situation based on valve rhythm behavior. **a** panel of fundamental information including water quality, ratio of simulation time, the occurrence time of transitional center in curve, time constant, As(V) activity coefficient, and present and integrated times. **b** built-in function of time-varying $n(t_R)$ and **c** $R_{max}(t_R)$ for As(V) induced valve response dynamics, **d** the profile of time-varying of input As(V) concentration and **e** the corresponding As(V) activity profile



between As(V) activity and dissolved As(V) is based on the water chemistry principle of activity coefficient (Eqs. 3 and 4).

The built-in database of $EC50_{As(V)}(t_R)$ was based on Eq. 5 and the interface of $EC50_{As(V)}(t_R)$ was presented in Fig. 3a. The site-specific $EC50_{\{As(V)\}}(t_R)$ can be accordingly obtained based on the As(V) activity coefficient calculated according to site-specific temperature and pH (Fig. 3b). Figure 3c demonstrates the interface of time-dependent dose-valve closure response profile ($R(t_R, As(V))$) based on Eq. T1 (Table 1). Figure 3d displays the interface of valve daily opening rhythm with site-specific As(V) exposure ($\psi(t, \{As(V)\})$) and unpolluted ($\psi(t, 0)$),

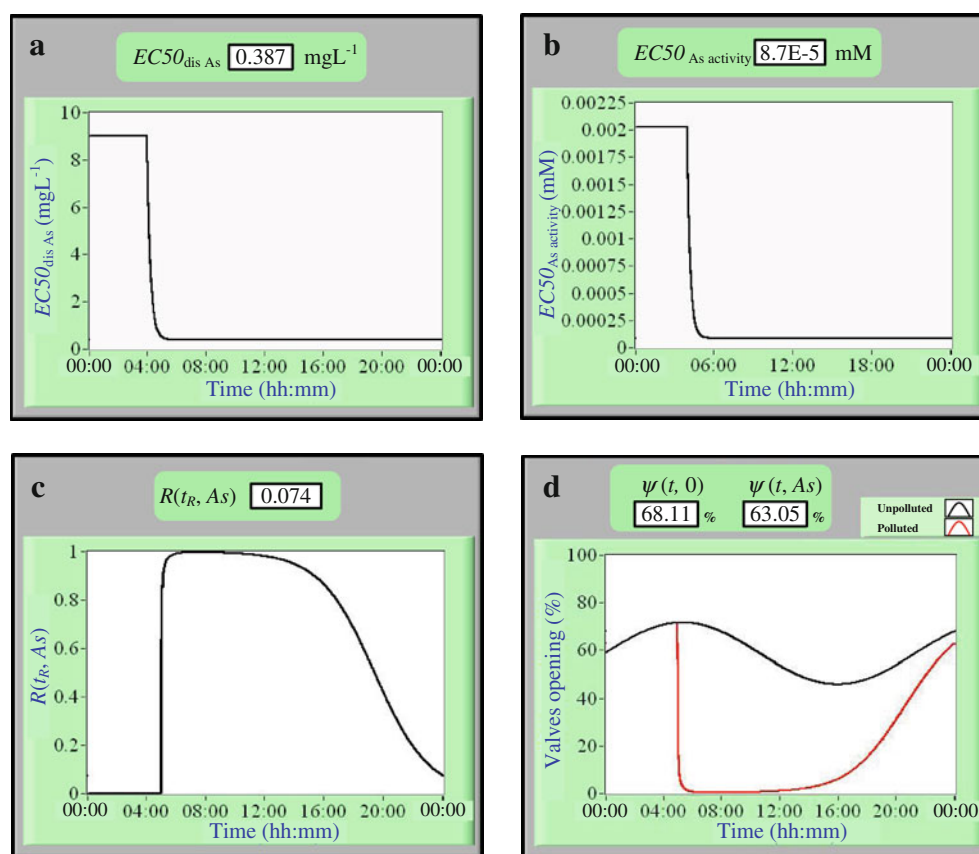
demonstrating the proportions of valve opening state with/without As(V) exposures, respectively.

Verification of biological early warning system

To further evaluate the practicability of the proposed online *C. fluminea*-based biomonitoring system, the experimental data of *C. fluminea* valve movement response to waterborne As(V) reported in previous study (Liao et al. 2009) were used for verification.

Here we performed five simulations in that the As(V) exposure concentrations were 0.3, 0.5, 1, 5 and 10 mg L⁻¹. The As(V) exposures were started at AM 3:00. Figure 4

Fig. 3 Monitoring interface showing the simulated dynamics of valve opening behavior in response to waterborne As(V). **a** built-in time-dependent EC50 profile for dissolved As, **b** time-dependent EC50 profile for As(V) activity, **c** time-dependent Hill model-based dose-valve closure response profile as the function of integration time, and **d** time-dependent valve daily opening rhythm with and without waterborne As(V) exposures



demonstrates the interfaces of valve daily opening rhythm during AM 03:00–08:00. The monitoring time duration was 30 min. These five interfaces displayed the percentage values of valve opening with/without As(V) exposure at AM 08:00. Our results indicated that valve opening percentage without As(V) exposure ($\psi(t, 0)$) was 67.77%, whereas $\psi(t, \{As(V)\})$ were 49.9, 24.5, 12.3, 0, and 0, respectively, in response to 0.3, 0.5, 1, 5 and 10 mg L⁻¹ As(V) (Fig 4a–e).

Given the measured $\psi(t, \{As(V)\})$ and built-in data $\psi(t, 0)$, the time-dependent valve closure response $R(t_R, \{As(V)\})$ can be displayed based on Eq. 1. The demonstrations of $R(t_R, \{As(V)\})$ in response to varied As(V) exposures were shown in Fig. 5. The measured $R(t_R, \{As(V)\})$ values at AM 08:00 were 0.26, 0.64, 0.82, 1, and 1, respectively, exposed to 0.3, 0.5, 1, 5 and 10 mg L⁻¹ As(V) (Fig. 5a–e).

The measured $R(t_R, \{As(V)\})$ shown in Fig. 5 could be used to obtain directly the site-specific As(V) activity and dissolved As (V) based on Eq. T1 (Table 1) in proposed five situations. Figure 6 displays the interfaces of time-dependent dissolved As(V) concentration. On the other hand, the As(V) activity can also be estimated to be 0.046, 0.121, 0.214, 0.367, and 0.374 nM in response to 0.3, 0.5, 1, 5 and 10 mg L⁻¹ As(V) at AM 08:00 (not shown).

Figure 6 also demonstrates the comparisons of the experimental data with the simulations. As shown in Fig. 6, the simulated dissolved As(V) concentrations were 0.21, 0.54, 0.95, 1.63, and 1.69 mg L⁻¹ at AM 08:00, respectively, at 0.3, 0.5, 1, 5 and 10 mg L⁻¹ As(V) exposure situations. We found that time-course changing profiles of dissolved As(V) derived from experimental $R(t_R, \{As(V)\})$ data were consistent with those in this study (Fig. 6a–c), except for the exposure As(V) concentrations higher than 1 mg L⁻¹ (Fig. 6d, e). This result implicated that As concentration detection limit of this proposed *C. fluminea*-based biomonitoring system should not be higher than 1 mg L⁻¹. Figure 6 also reveals that the detection times are much dependent on the As(V) exposure concentrations.

Discussion

Detection limitation

This study synthesized a real-time biomonitoring system to detect As(V) toxicity by valve movement of *C. fluminea* in response to 0, 0.3, 0.5, 1, 5, and 10 mg L⁻¹ As(V). Our study revealed that when *C. fluminea* exposed to 1, 5, and 10 mg L⁻¹ As(V) longer than 1 h, the valve closure responses were

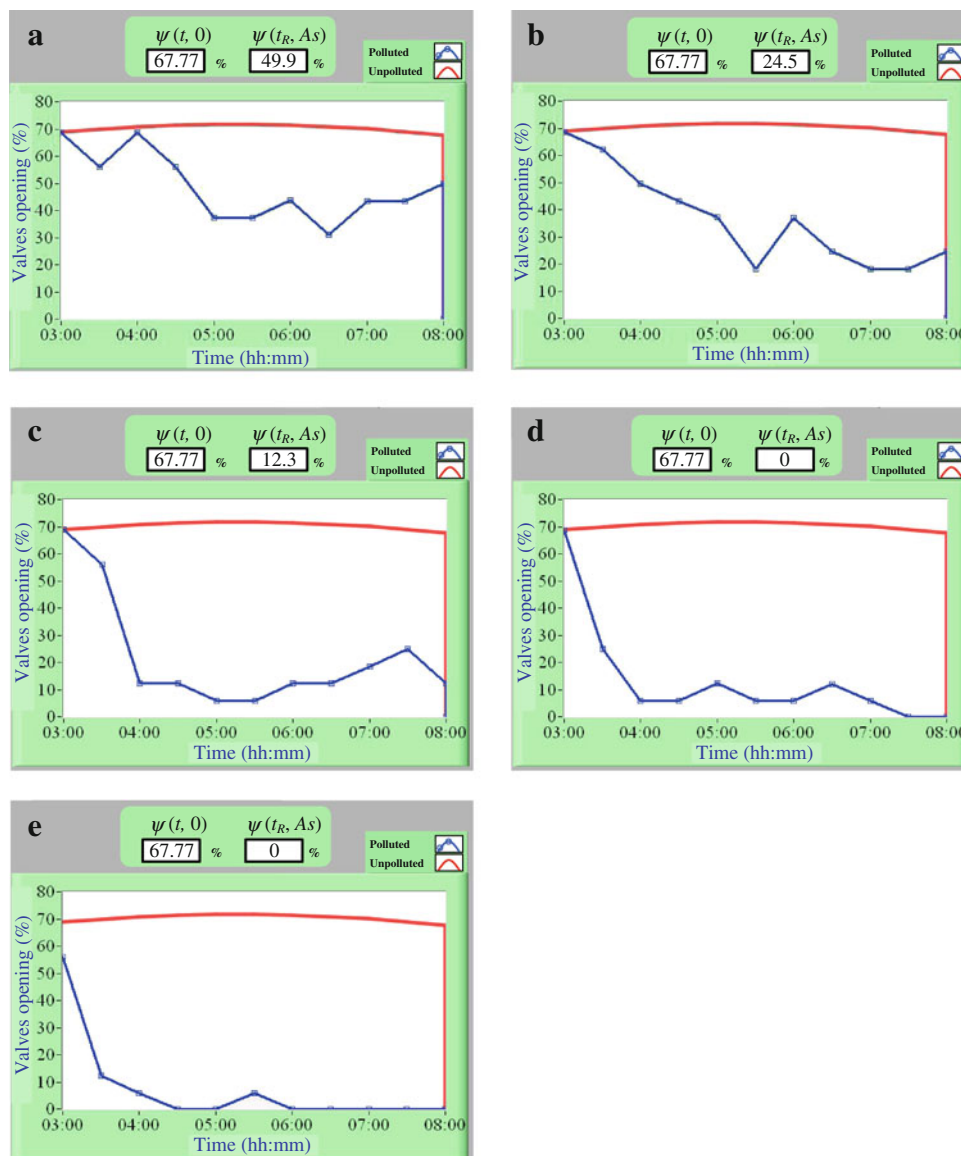


Fig. 4 Monitoring interface showing the real-time observed clam valve daily opening behaviors in response to waterborne As(V) concentrations of **a** 0.3, **b** 0.5, **c** 1, **d** 5, and **e** 10 mg L⁻¹, respectively,

achieved more than 80% closure responses. These response signals cannot reveal the significant differences among waterborne As(V) of 1, 5, and 10 mg L⁻¹ in inducing valve closure behaviors. The maximum valve closure response merely reflects 1.69 mg L⁻¹ waterborne As(V).

The applicability of detection has been verified with the published experiment data. Practically, the performance of this proposed biomonitoring system demonstrates fairly good applicability to detect waterborne As concentrations when the field As concentrations are less than 1 mg L⁻¹. The valve dynamic monitoring system presented the better predictive As concentration capabilities that should be limited at 1.69 mg L⁻¹. The results of this proposed

based on published experimental data (Liao et al. 2009). The clam valve daily opening profile without As(V) exposure is also shown (*red line*) (color figure online)

biomonitoring system met this principle, since 0–1 mg L⁻¹ of experimental exposure concentrations confirmed the polluted arseniasis-endemic area in Taiwan where 0.04–0.9 mg L⁻¹ of As in surface waters were found (Lin et al. 2001, 2005; Liao et al. 2003; Huang et al. 2003; Liu et al. 2005, 2006, 2007).

Tran et al. (2004) indicated that the detection times were associated with the exposure concentrations. The high concentration exposure should be monitored in the shorter detection times. The results of this study were agreed with this principle. Since the aquatic organisms merely absorb slightly the exposure concentration within a short period of time that did not cause the fierce valve closure reaction,

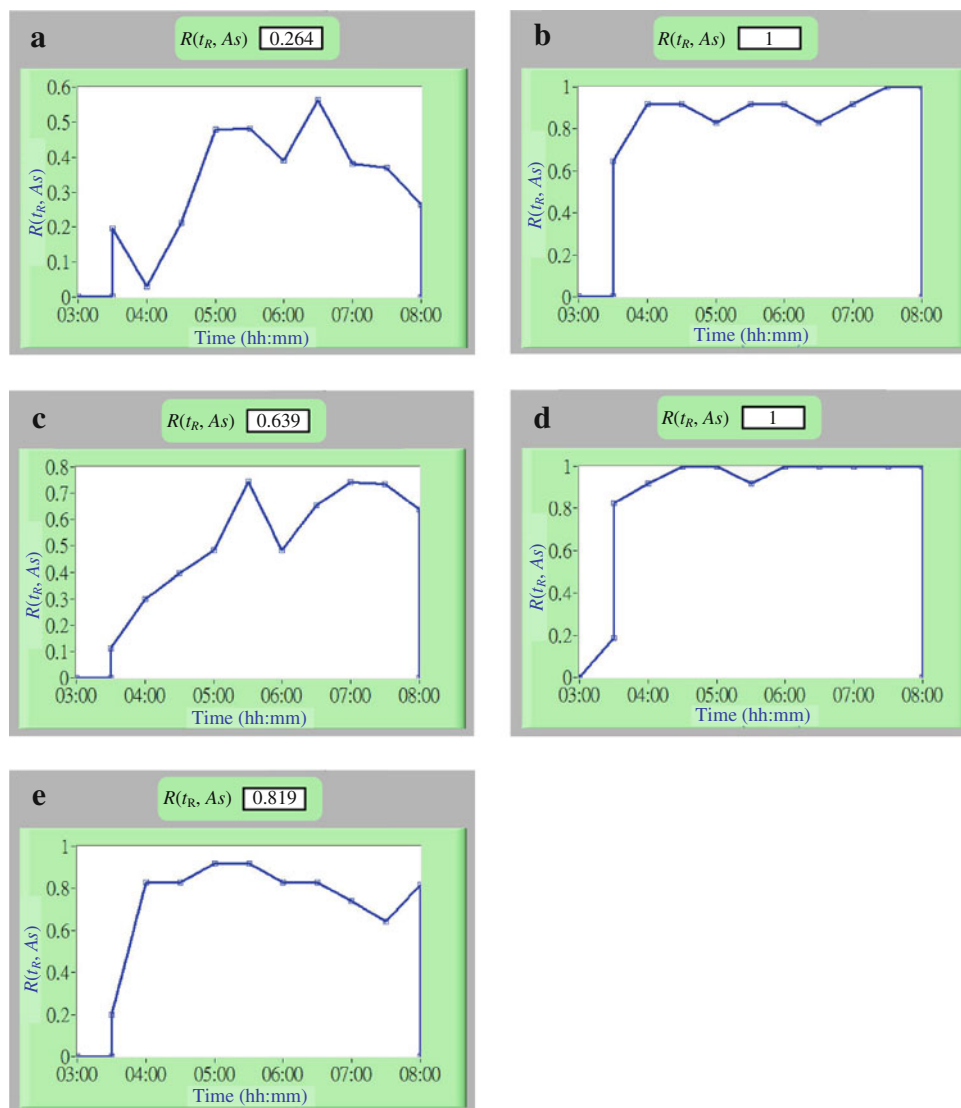


Fig. 5 Monitoring interface illustrating the real-time valve closure response profiles in response to waterborne As(V) concentrations of **a** 0.3, **b** 0.5, **c** 1, **d** 5, and **e** 10 mg L^{-1} , respectively, converted from Fig. 4

whereas the high concentration induced valve closure reactions are nearly corresponding to that of low concentration exposure in the long detection time. The detection threshold could be obtained from Eq. 5 to reflect the response time of valve closure in As polluted surface waters. Therefore, detection thresholds of 5 and 0.5 mg L^{-1} As are <9 and <62 min, respectively.

The valve rhythm activity in *C. fluminea* can be used to develop the biological early warning system to monitor water quality. Behavior signals in ecotoxicology could provide the bio-effect related monitoring for chemical toxicity (Alcock et al. 2003). This early warning system could display unknown chemical compounds concentration by the response signals. The detection program should be made with the sensitivity of aquatic organism behavior

close to the exposure chemical concentrations in the aquatic ecosystem (Hansen 2008). The uncertainty of valve movement detection could also cause misestimates that should be taken into account in evaluating the waterborne As concentration in the field aquatic environments.

Various environmental conditions

Previous studies indicated that many abiotic factors affect the behaviors of aquatic organism, such as water temperature, tides, photoperiod, food abundance, turbidity, and dissolved oxygen concentration (Anestis et al. 2007; Chambon et al. 2007; Soucek 2007; Bacci et al. 2008; Gnyubbkin 2010). The valve rhythm of bivalve depends on the environmental variations. The observed behavior of

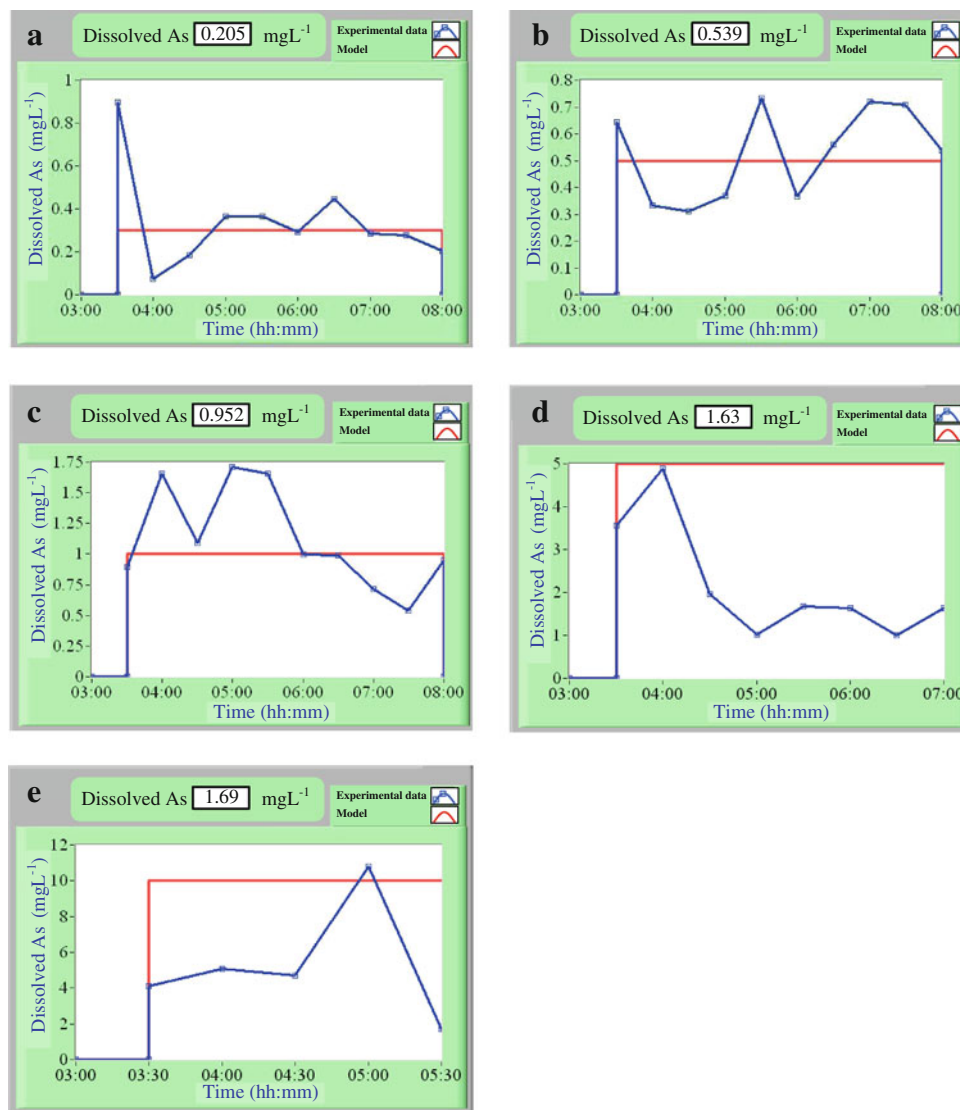


Fig. 6 Performance and results of the system verification based on the designed window of a monitoring interface showing the comparisons between the measured experiment data (blue line) and the simulated

dissolved As(V) concentrations (red line) based on the proposed *C. fluminea*-based biomonitoring system in the As(V) exposure conditions of **a** 0.3, **b** 0.5, **c** 1, **d** 5, and **e** 10 mg L⁻¹ (color figure online)

C. fluminea and *Mytilus galloprovincialis* are related to their response to water temperature changes. Ortmann and Grieshaber (2003) indicated that valve opening of *C. fluminea* in the afternoon and valve closure during the night period at the water temperature ranged from 19.1 to 22.4°C, whereas valve closure could remain for the longer periods at water temperature below 5°C. For *M. galloprovincialis*, the water temperature increased from 7 to 16°C and the valve closure behavior would be decreased, whereas the valve closure increased as the water temperature increased to 24°C (Anestis et al. 2007).

Moreover, the turbidity in aquatic environments could induce the direct harmful effects on bivalve. The bivalve reduces the feeding activity and efficiency of gas exchange

to regulate the metabolic mechanism (Bacci et al. 2008). These behavior activities are directly related to the degree of valve gaping. Moreover, Gnyubbkin (2010) investigated the effect of photoperiod to the valve daily rhythm of *M. galloprovincialis*, indicated that the stimulation of dark lighting induced the immediate valve closure behavior even in a few second. Bivalves take the dark light to be the approach of predators and close the valves to protect themselves. A plenty of environmental conditions like photoperiod, water temperature, and tide are also displaying the seasonal rhythm. Previous studies indicated that seasonal temperatures (summer and winter) could directly affect the valve rhythm behaviors (Ortmann and Grieshaber 2003; Anestis et al. 2007).

Response signal with physiological variation

Previous studies used numerous different behaviors of bivalve to observe the physiological rhythm such as metabolism rate, cardiac function, mantle movement, siphon extension, byssus production, burrowing behavior and valve movement (Ortmann and Grieshaber 2003; Bakhmet and Khalaman 2006; Rodland et al. 2006; Bonnard et al. 2009; Jou et al. 2009; Liao et al. 2009). The signal signatures are species and size dependent. The valve activity is related to the physiological process such as feeding, respiration, and metabolism. Ortmann and Grieshaber (2003) indicated that *C. fluminea* reduced its energy metabolism rate to 10% below the base metabolism rate when valve closure. These measured metabolism rate were based on the heat dissipation and oxygen consumption endpoint. The strong metabolism rate of aquatic organism was dependent on the faster breathing, whereas the lower metabolism rate reflected the slow ventilation rate. These variations of physiological factors could also affect the valve closure behavior.

More generally, valve behavior is related to the relaxing of the adduction and thus to passive valve opening. Fdil et al. (2006) indicated that some valve opening state of bivalve was dependent on the mantle edges extension and the siphons opening and protruding to visual observe the valve rhythm in the normal activity. The observed frequency and magnitude of valve gap were not suitable as the toxic effect endpoint alone. Hence, it is important to enhance the applicability of biomonitoring technique by taking siphons extension and valve gap into account for more accurate and robust detection of pollutant exposure concentrations, and holds a high promise for further application.

In conclusion, this study provides a parsimonious methodology to synthesize a real-time biomonitoring system based on *C. fluminea* valve rhythm behavior and to detect the site-specific waterborne As exposure. On the other hand, the LabVIEW graphic control program based biomonitoring system allows a rapid and cost-effective method to implement the online As detection. The core of this biomonitoring system is computational mechanisms I and II, clam closure response behavior and Hill-based dose-response model, which offer the basis for a quantitative and comprehensive assessment of clam-based BEWS. The great accuracy and sensitivity of the two mechanisms enable a reliable early warning of potential risks posed by toxic level of As. Their relative simplicity also ensures a high possibility for being incorporated into an automated control system. In the future work, the detect mechanisms should include the seasonal photoperiods- and dissolved oxygen-dependent normal rhythm integrated with valve rhythm and adduction of rhythm mechanisms to perform the sensitivity in early warning biomonitoring system.

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