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A flow-through passive dosing system for continuously supplying aqueous solutions of hydrophobic chemicals to bioconcentration and aquatic toxicity tests

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ABSTRACT

A continuous supply of water with defined stable concentrations of hydrophobic chemicals is a requirement in a range of laboratory tests such as the OECD 305 protocol for determining the bioconcentration factor in fish. Satisfying this requirement continues to be a challenge, particularly for hydrophobic chemicals. Here we present a novel solution based on equilibrium passive dosing. It employs a commercially available unit consisting of ~16 000 polydimethylsiloxane (PDMS) tubes connected to two manifolds. The chemicals are loaded into the unit by repeatedly perfusing it with a methanol solution of the substances that is progressively diluted with water. Thereafter the unit is perfused with water and the chemicals partition from the unit into the water. The system was tested with nine chemicals with log K_{OW} ranging from 4.1 to 6.3. The aqueous concentrations generated were shown to be largely independent of the water flow rate, and the unit to unit reproducibility was within a factor of ~2. In continuous flow experiments the aqueous concentrations of most of the study chemicals remained constant over 8 d. A model was assembled that allows prediction of the operating characteristics of the system from the log K_{OW} or PDMS/water partition coefficient of the chemical. The system is a simple, safe, predictable and flexible tool that generates stable aqueous concentrations of hydrophobic chemicals.

1. Introduction

The generation and maintenance of defined concentrations of hydrophobic organic chemicals in large quantities of water is important in different areas of contaminant science. In experimental protocols to study fish bioconcentration such as the OECD 305 guideline (OECD, 1996), stable concentrations must be maintained over a period of several weeks. Similarly, constant exposure concentrations are often required for chronic toxicity testing of aquatic macrofauna such as fish (ECETOC, 1996; OECD, 2000). These experiments must generally be conducted under flow-through conditions, which requires a system that can continually produce contaminated water for several weeks.

Scientists have been developing solutions for this problem for many decades. Mount and Brungs (1967) presented a system made out of readily accessible materials that diluted saturated slurries of pesticides and delivered them continuously over several weeks to fish exposure experiments. When research expanded to chemicals that are sparingly soluble in water, generator columns were introduced to generate saturated aqueous solutions, which were subsequently diluted (Muir et al., 1986). Others have chosen to use solubilising agents (Yakata et al., 2003, 2006) or slow stirring (for liquids that are immiscible with water) (Tolls and van Dijk, 2002) to create solutions of sparingly soluble chemicals, which are then diluted. For more water soluble substances methanol solutions of the test chemical have been directly diluted with water (Smith and Hill, 2004). Many of these methods are cumbersome, difficult to implement, or can raise occupational health concerns due to the need to handle large quantities of the test substance. Generating stable aqueous concentrations of hydrophobic chemicals continues to be a technical obstacle for the use of flow-through test protocols, and hence scientists continue to use less desirable exposure protocols such as static exposures with solution replacement (e.g. Paterson and Metcalfe, 2008).

The equilibrium passive dosing techniques developed in recent years may represent a viable alternative to existing methodologies. In equilibrium passive dosing, the exposure medium is brought into contact with another phase containing a significant reservoir of the chemical (Mayer et al., 1999). The chemical partitions from the reservoir into the medium until a partitioning equilibrium is approached. If chemical is added to or removed from the medium by another process, then this chemical partitions to achieve a new equilibrium between the medium and the reservoir. Since the vast majority of the chemical is present in the reservoir, it buffers these disturbances, and the concentration in the medium is maintained constant. This methodology is particularly suitable for

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hydrophobic chemicals, as this facilitates the creation of reservoirs with a storage capacity much greater than water. Typically polymers such as the silicone polydimethylsiloxane (PDMS) are used, and PDMS has been successfully employed for passive dosing in laboratory toxicity tests of small organisms, fish embryos and cell cultures using test vessels of 1–100 mL (Brown et al., 2001; Kiparissis et al., 2003; Mayer and Holmstrup, 2008; Bandow et al., 2009; Smith et al., 2010). The PDMS can easily be applied at the bottom or on the vertical walls of such smaller test systems to provide a sufficient surface for an efficient passive dosing of aqueous concentrations. This is not possible in larger test systems such as aquaria of tens to hundreds of litres. Within other technical areas, hollow fibre technology is then often applied to provide markedly higher surface areas. This has already been utilised for equilibrium sampling techniques using hollow fibre modules (Larsson et al., 2009) and also PDMS hollow fibres (Mayer et al., 2009).

In this paper we explore the use of a commercially available hollow fibre module for flow-through passive dosing of large volumes of aqueous solutions. This device combines a large quantity of PDMS with a very large PDMS/liquid contact area which facilitates partitioning equilibration between the chemical reservoir in the PDMS and the flowing liquid. The ability of the system to produce constant and repeatable aqueous concentrations over weeks of exposure was tested for nine chemicals covering a two orders of magnitude range of hydrophobicity.

2. Methods

2.1. Passive dosing unit

A PermSelect® PDMSA-1.0 silicone membrane module (MedArray, Ann Arbor, MI, USA) was used for the passive dosing (Fig. 1). It was 5 cm in diameter, 14 cm long, weighed 180 g, and consisted of two manifolds connected by hollow fibres. According to the manufacturer there were 16128 fibres with an inner and outer diameter of 167 μm and 237 μm, respectively. The tubes were approximately 11 cm long, giving an estimated total PDMS volume of 39.4 mL and total PDMS mass of 38 g. The total internal surface area was given as ~1 m^2, which is orders of magnitude higher than previously published passive dosing formats with typical contact areas of 1–10 cm^2 (Mayer et al., 1999; Brown et al., 2001; Bandow et al., 2009; Smith et al., 2010). The unit offered the possibility to perfuse both the inner (lumen) and the outer (shell) surface of the tubes. The housing and fittings were made of polycarbonate, while the interior of the device contained some polypropylene and acrylic resins.

Fig. 1. The passive dosing unit showing the inlet (lower left) and outlet (lower right) ports for the perfusion of the lumen. The other three ports are for the perfusion of the shell space.

2.2. Chemicals, solvents and sorbents

Ethyl acetate ('Pestiscan') was purchased from Labscan Ltd., Dublin, Ireland; methanol ('LiChrosolve') from Merck Darmstadt, Germany; SPE cartridges (1 mL 50 mg ISOLUTE ENV+) from Biotage AB, Uppsala, Sweden. The water used for diluting the standard solutions was of milli-Q grade from a milli-Q ultrapure water system, MilliQ PLUS 185 from Millipore Stockholm, Sweden. Chlorpyrifos, 2,3,4-trichloroanisole, pentachlorobenzene, and 4-n-nonylphenol were from Honeywell Specialty Chemicals, Seelze, Germany; 2,6-di-*tert*-butylphenol, and 2,4,6-tri-*tert*-butylphenol from Sigma–Aldrich, Schnelldorf, Germany; 2,5-dichlorobiphenyl from Accustandard, Inc., New Haven, CT, USA; 2,4,5-trichloroanisole from Ultra Scientific, Inc., North Kingstown, RI, USA; p,p′-DDT and D15-Musk xylene from Dr. Ehrenstorfer, Augsburg, Germany; $^{13}C_{12}$-2,5-dichlorobiphenyl, $^{13}C_{12}$-4,4′,DDT, $^{13}C_6$-hexachlorobenzene, $^{13}C_6$-4-n-nonylphenol, $^{13}C_6$-pentachlorobenzene and diethyl-D10 –chlorpyrifos from Cambridge Isotope Laboratories, Inc., Andover, MA, USA; decachlorobiphenyl from Wellington Laboratories, Guelph, ON, Canada.

2.3. Loading and operation of the passive dosing unit

The test chemicals (2,3,4-trichoroanisole, musk xylene, 4-n-nonylphenol, chlorpyrifos, pentachlorobenzene, 2,4,6-tributylphenol, 2,5-dichlorobiphenyl, hexachlorobenzene and p,p′-DDT) were selected to cover a range of hydrophobicity. Their logK_{OW} values are given in Table S1 (Supplementary material).

A loading solution of the test chemicals was prepared in 25 mL of methanol (Table S1 lists the mass of each chemical used in experiment #1). The loading solution was introduced into the fibre lumen inlet port of the passive dosing unit and allowed to stand for 30 min. The unit was tipped, and the solution drained out of the fibre lumen outlet port and was collected. Nitrogen was used to displace any remaining solution. Water (2 mL) was added to the solution, and the solution was re-introduced into the inlet port, displaced with air after several minutes, and recollected. This procedure was repeated nine times using water dilution volumes of 4, 8, 8, 10, 10, 25, 50, 100, and 250 mL, so that the final injection was 492 mL of a 5:95 (v:v) methanol:water mixture. The larger volumes were perfused at ~100 mL min^{-1} through the unit. The successive dilution of the loading solution with water encouraged the partitioning of the chemicals into the dosing unit (Birch et al., 2010).

The fibre lumen inlet port was then connected to the water supply of the laboratory's aquaria facility. The water temperature was 13 °C. Water was allowed to flow through the unit at between 25 and 525 mL min^{-1}. The water flow was regulated by a clamp on the tube leading into the aquarium. The pre-loaded chemicals partitioned from the unit into the water. The ports to the shell volume of the passive dosing unit were sealed and not perfused.

2.4. Sampling and analysis of water

The water samples were collected directly in the elution tubes used for the SPE system (70 mL, polyethylene, with a stopcock). A solution of surrogate standard in methanol was prepared. The surrogate standard used for each analyte and its concentrations in the surrogate standard solution are given in Table S1 (Supplementary material). A 2 mL portion of the surrogate standard solution was added to the SPE elution tube. The water sample (~60 mL) was collected by holding the tube under the outlet port of the passive dosing unit or submerging one side of the lip of the tube in the aquarium. The mass of the tube was recorded before and after water sampling to determine the mass of the sampled water. The sample was transferred to a 50 mg ENV + SPE cartridge, which

was dried with nitrogen, after which the analytes and surrogate standards were eluted with 1 mL of ethyl acetate containing 40 ng of decachlorobiphenyl as a volumetric standard and analysed without further treatment.

Gas chromatographic separation was performed on a CarloErba GC8000 gas chromatograph using a 30 m × 0.25 mm i.d. DB-5MS J&W Scientific column with a 0.25 μm film-thickness. Injections were made in the splitless mode (260 °C) with the GC oven at 80 °C. This was held for 2 min, raised to 300 °C at a rate of 10 °C min^{-1}. Mass spectrometric detection was made using a Finnigan Voyager low resolution mass spectrometer in electron impact mode. The method detection limit ranged from 5 to 20 ng L^{-1} for the different analytes.

2.5. Experiments

Experiment #1 tested the reproducibility of the loading of the passive dosing system and the effect of flow velocity on the analyte concentrations in water leaving the unit. Three passive dosing systems (denoted PS2, PS4, and PS6) were loaded in an identical manner with the chemical quantities given in Table S1 (Supplementary material). The units were connected to the water supply system. The water flow through the units was adjusted to approximately 30 mL min^{-1}. After 1 h to allow purging of residual methanol from the system, three water samples were collected from the outlet port of each unit. The procedure was repeated at water flows of 200 and 500 mL min^{-1}.

Experiments #2–4 tested the ability of the system to produce constant concentrations in aquaria over longer periods of time. In experiment #2 two passive dosing units that had each been loaded with sub-sets of the chemicals were connected to a 200 L aquarium made of fibreglass. The water flow through each of the passive dosing units into the aquarium was set to 200 mL min^{-1}. A chemical free water supply was used to maintain the total water flow through the aquarium at 800 mL min^{-1}, providing a water exchange rate of once every 4 h. A water sample was collected from the drain of the aquarium at six time points over 8 d. Experiments #3 and #4 were similar, except that the aquarium now contained fish (20–60 rainbow trout, ~45 g, fed daily 1% of body weight). In both experiments the water flow through the passive dosing units and the aquarium was 200 and 800 mL min^{-1}, respectively. This corresponded to a fish loading rate of 0.8–2.3 g fish per L d^{-1} of water flow through, which is at the upper end of the recommended range in the OECD 305 guideline for measuring fish bioconcentration factors (OECD, 1996). Only the three chemicals with lowest K_{OW} which were depleted in the passive dosing system most quickly, namely 2,3,4-trichloroanisole, musk xylene and chlorpyrifos, were quantified in these two experiments.

Finally, a simple test was conducted to assess whether it is possible to store passive dosing units that had been loaded with chemicals, or whether they have to be freshly loaded. A unit that had been loaded and then perfused for 14 d was drained of water and stored at room temperature for 6 months, after which it was reconnected to the aquarium system. The concentrations of 2,3,4-trichloroanisole, musk xylene and chlorpyrifos measured in the aquarium water during the 3 d prior to taking the unit out of service were compared with the concentrations measured during the 3 d after it was started up again.

3. Results and discussion

3.1. Reproducibility of the loading and dosing

Table 1 summarises the results of the reproducibility testing of the loading procedure. The concentrations produced by the passive dosing units PS2 and PS4 were each normalised to the concentrations produced by the unit PS6, whereby the ratios were calculated from the mean concentrations for a given flow velocity, and then the ratios were averaged for a given compound across the three flow velocities. The mean ratios ranged between 0.76 and 2.31. With the exceptions of 4-n-nonylphenol and musk xylene, the concentrations of all substances were highest in PS2, followed by PS4. This suggests that the differences between the units were largely independent of substance properties.

Table 1
Reproducibility of the loading of the passive dosing unit, expressed as the quotient of the chemical concentration in water in units PS2 and PS4 normalised to the concentration from PS6.

	PS2/PS6	PS4/PS6
2,3,4-Trichloroanisole	1.70	1.35
Musk xylene	1.00	0.84
Chlorpyrifos	1.75	1.32
4-n-nonylphenol	1.16	0.76
2,4,6-Tri-*tert*-butylphenol	1.38	1.18
Pentachlorobenzene	1.43	1.22
2,5-Dichlorobiphenyl	1.82	1.56
Hexachlorobenzene	1.59	1.33
p,p′-DDT	2.31	1.18

There are a number of potential causes of variability in the loading procedure. One is variability in the extent of transfer of the chemicals from the loading solution to the PDMS. However, analysis of the loading solution after the loading procedure showed that <0.5% of the chemicals remained in the solution, suggesting that transfer to the PDMS was nearly complete. A second explanation is that there were differences between the silicone mass in the different units, but inspection of the units indicated that it was unlikely that the variability could be as large as a factor 2. As the results of the modelling (see below) suggested that the dominant sorbent in the units was not silicone, a third possibility is that there were differences in the quantities of other materials in the units. The causes of the variability were not explored further, as the observed level of reproducibility was considered sufficient for common applications of large volume passive dosing to water, such as fish bioconcentration experiments.

3.2. Influence of flow rate

The influence of the water flow rate on the concentrations in water is shown for 2,3,4-trichloroanisole, musk xylene, 2,5-dichlorobiphenyl and p,p′-DDT in Fig. 2, while the results for the remaining substances are presented in Fig. S1. For a given passive dosing unit, the influence of water flow rate was generally <20%. The differences between the three flow rates were largely consistent between the three units tested. No consistent trend of increasing or decreasing concentration with flow rate was observed for most of the chemicals. The exception was p,p′-DDT, for which the concentrations were lowest at the lowest water flow rate. This is contrary to what would be expected should the PDMS-to-water mass transfer be too low to allow the equilibration of chemicals between the tubing and the water during its passage through the unit. The absence of a marked influence of flow rate on test chemical concentration means that the flow rate through the passive dosing unit can be used to regulate the influx of solution into the test system (e.g. aquarium). The dynamic range of this regulation is at least a factor of 20.

3.3. Constancy of the chemical concentrations in water

The chemical concentrations in water sampled from the drain of the aquarium during experiment #2 are plotted in Fig. 3 for 2,3,

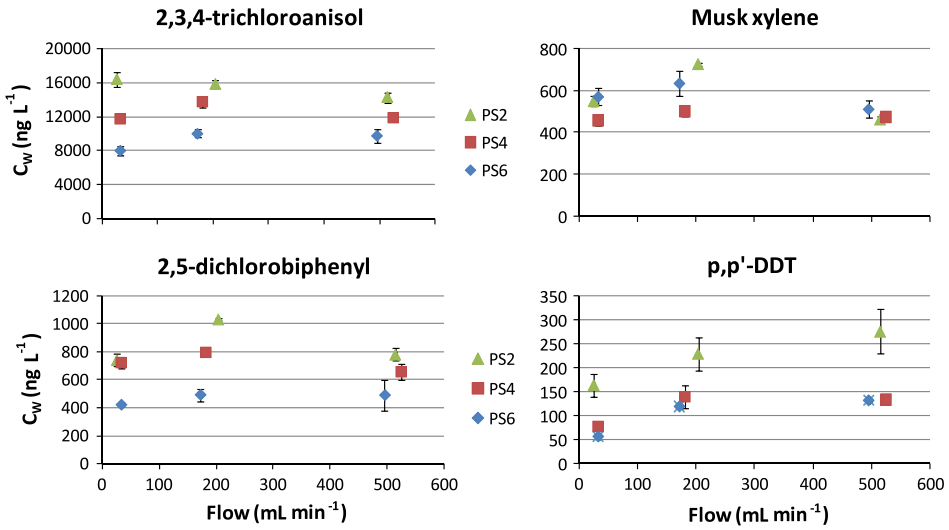

Fig. 2. Influence of flow rate on the concentrations of 2,3,4-trichloroanisole, musk xylene, 2,5-dichlorobiphenyl and p,p′-DDT in water. The symbols distinguish the results for three different passive dosing units that were loaded in the same manner. The error bars represent the standard deviation of three water samples.

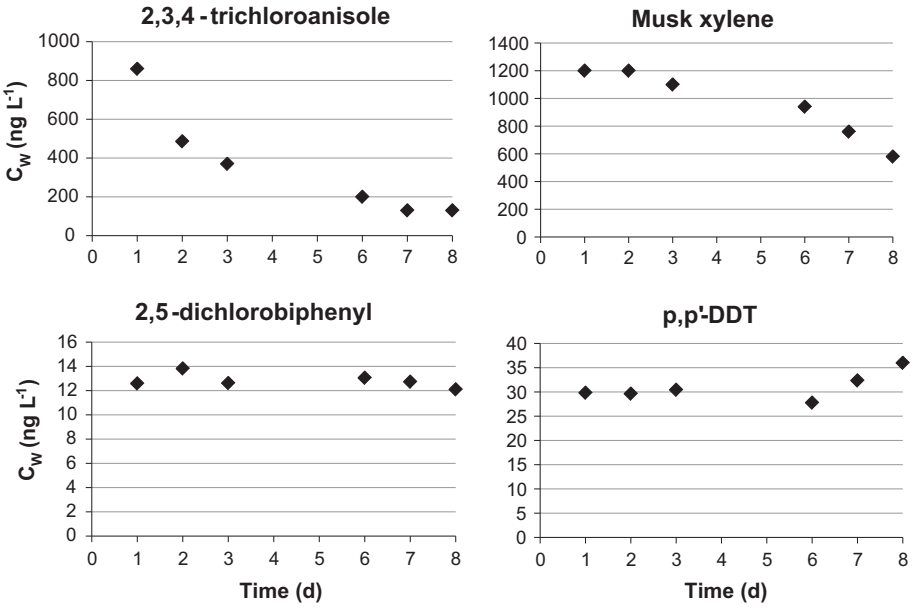

Fig. 3. Concentrations of 2,3,4-trichloroanisole, musk xylene, 2,5-dichlorobiphenyl and p,p′-DDT in aquarium water, generated during continuous operation of the passive dosing system at 200 mL min^{-1}. The aquarium did not contain fish (Experiment #2).

4-trichloroanisole, musk xylene, 2,5-dichlorobiphenyl and p,p′-DDT, while the results for the remaining substances are shown in Fig. S2. The concentrations of the hydrophobic chemicals (pentachlorobenzene, 2,5-dichlorobiphenyl, 4-n-nonylphenol, p,p′-DDT, hexachlorobenzene, and 2,4,6-tri-*tert*-butylphenol) showed no consistent trend over time. The relative standard deviations of the measured concentrations were 9%, 5%, 12% and 9% for the first four chemicals, indicating that the passive dosing system produces constant concentrations of these substances. The relative standard deviations of hexachlorobenzene (25%) and 2,4,6-tri-*tert*-butylphenol (27%) were higher. No explanation for their poorer reproducibility was found.

The concentrations of the more hydrophilic chemicals (2,3,4-trichloroanisole, chlorpyrifos, and musk xylene) showed a decreasing trend with time. This was also observed for these three chemicals in experiments #3 and #4 in which the units were operated continuously for 13 and 17 d (Fig. 4). Although the decrease in these two experiments amounted to 13–85% over the exposure period, the concentrations were relatively constant from day to day (median day to day variability 13%, 6% and 11%).

3.4. Long-term stability of the system

After a passive dosing unit had been stored for 6 months and then put back into operation using the same water flow, the concentration of chlorpyrifos (507 ng L^{-1}, average over 3 d) in the water draining the aquarium was very similar to prior to storage (493 ng L^{-1}, average over 3 d). For 2,3,4-trichloroanisole and musk xylene the concentrations were somewhat higher after storage than before (570 versus 310 and 1170 versus 774 ng L^{-1}, respectively). These are the two chemicals that show the strongest depletion during long term operation of the passive dosing units.

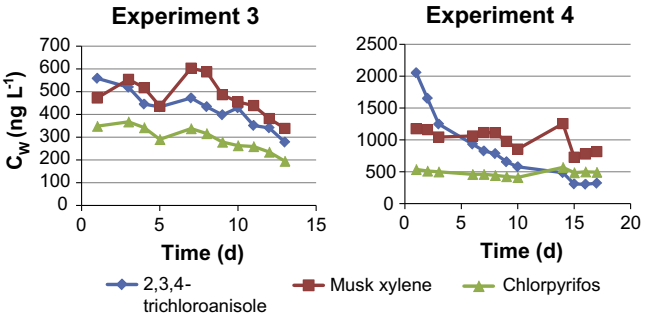

Fig. 4. Concentrations of 2,3,4-trichloroanisole, musk xylene and chlorpyrifos in aquarium water, generated during continuous operation of the passive dosing system at 200 mL min^{-1} for 13 and 17 d. The aquaria contained fish.

The results suggest that there is a small recovery effect during storage, perhaps as a result of redistribution of the chemical within the passive dosing unit. Overall the results suggest that a loaded unit can be stored for several months without this having a large impact on the concentrations generated.

3.5. Mathematical model of the passive dosing system

In order to garner mathematical understanding of the system, dosing system/water partition coefficients ($K_{ds/w}$) were calculated from the results of Experiment #1. The concentration in the dosing system (C_{ds}, mol unit^{-1}) was estimated by dividing the mass of chemical in the dosing solution by one unit, and this was in turn divided by the concentration in water (C_W, mol L^{-1}) collected at the outlet port of the unit. $K_{ds/w}$ has units of L unit^{-1} and indicates the volume of water that would contain the same amount of chemical as the unit at equilibrium. This procedure assumes that all of the chemical loaded into the system partitions into the unit, and that a partitioning equilibrium was achieved between the unit and the water. The former was demonstrated by the small fraction of chemical left in the loading solution following loading, while the latter is a good assumption, given that the concentrations of the chemicals in the water were largely independent of the water flow rate.

The experimental $K_{ds/w}$ values are compared with $K_{PDMS/w}$ values from the literature in Table 2. When available, measured $K_{PDMS/w}$ values were taken from the literature, while for the remaining chemicals $K_{PDMS/w}$ was predicted from the chemical's octanol–water partition coefficient using the regression of DiFilippo and Eganhouse (2010) (see Table 2). There was a good correlation between $\log K_{ds/w}$ and $\log K_{PDMS/w}$ ($r^2 = 0.83$).

The kinetics of chemical depletion in the passive dosing system was evaluated using a one compartment model and assuming that elimination was a first order process. The concentration in water as a function of time $C_W(t)$ was described using:

$$C_W(t) = C_W(0)e^{-kt} \quad (1)$$

where $C_W(0)$ is the initial concentration in water (g L^{-1}), t is time elapsed since the initial concentration was measured (h), and k, the rate constant for elimination (h^{-1}), is defined by

$$k = \frac{q}{K_{ds/w}} \quad (2)$$

where q is the water flow rate through the dosing unit (L h^{-1}). Using this equation, modelled values of the rate constant k were calculated from the experimental $K_{ds/w}$ reported in Table 2 and the water flow rate of 13 L h^{-1} (constant for experiments 2, 3 and 4). For those chemicals for which the concentrations clearly decreased with time in experiments 2, 3 and 4 (2,3,4-trichloroanisole, chlorpyrifos, and musk xylene), experimental k values were calculated from semilogarithmic plots of the concentration in water against time (Table 3, note that the 95% confidence interval of the k value does not intersect zero in any case, confirming that there was a decrease in concentration over time). These experimental k values agree reasonably well with the modelled k values based on the experimental $K_{ds/w}$ (Table 3). Thus, the experimental $K_{ds/w}$ values are largely consistent with the observed depletion kinetics in the passive dosing system. This indicates that it is reasonable to model the system using a one compartment model and assuming that elimination was a first order process.

Table 2
Log PDMS/water partition coefficients ($K_{PDMS/w}$, literature), dosing unit/water partition coefficients ($K_{ds/w}$, measured), and partitioning capacity of the PDMS in the unit (estimated from $K_{PDMS/w} \times V_{PDMS}$) for the study chemicals.

	Log $K_{PDMS/w}$[a] [L L^{-1}]	Log $K_{ds/w}$ [L unit^{-1}]	Log ($K_{PDMS/w} \times V_{PDMS}$) [L unit^{-1}]
2,3,4-Trichloroanisole	3.47[b]	3.05	2.07
Musk xylene	3.72[b]	4.10	2.31
Chlorpyrifos	4.53	4.20	3.12
4-n-nonylphenol	5.13[b]	4.48	3.73
2,4,6-Tri-*tert*-butylphenol	4.47[b]	4.13	3.06
Pentachlorobenzene	4.39	4.17	2.99
2,5-Dichlorobiphenyl	4.40[b]	4.28	3.00
Hexachlorobenzene	4.91	4.56	3.51
p,p′-DDT	5.66	5.28	4.26

[a] Mean of the measured values that passed the quality assurance screen in DiFilippo and Eganhouse (2010). In the absence of measured values, the predictive equation $\log K_{PDMS/w} = 0.83 \log K_{OW} + 0.07$ from the same source was used.
[b] Predicted value.

Table 3
First order rate constants (h^{-1}) for the decrease in chemical concentration in water as measured in experiments 2, 3 and 4 (k (exp), mean ± standard error) and as predicted from the observed $K_{PDMS/w}$ value (k (mod)).

	k (exp 2)	k (exp 3)	k (exp 4)[a]	k (mod)
2,3,4-Trichloroanisole	0.0109 ± 0.0010	0.0019 ± 0.0003	0.0055 ± 0.0003	0.0115
Musk xylene	0.0039 ± 0.0007	0.0010 ± 0.0005	0.0009 ± 0.0004	0.0010
Chlorpyrifos	0.0016 ± 0.0006	0.0018 ± 0.0003	0.0011 ± 0.00006	0.0008

[a] The last four data points in the experiment were not used as a step increase in concentration suggested calibration error.

In choosing the PermSelect® unit for this application, it had been assumed that the PDMS tubes would provide the principle reservoir into which the hydrophobic chemicals would partition. This hypothesis was tested by comparing the measured partitioning capacity of the unit (i.e., $K_{ds/w}$) with the partitioning capacity of the PDMS in the unit, which was estimated as the product of $K_{PDMS/w}$ and the volume of PDMS in the unit. In all cases the partitioning capacity of the PDMS was much lower than the partitioning capacity of the unit, generally by more than an order of magnitude (Table 2).

One possible explanation for the lower estimated partitioning capacity of the PDMS is the low temperature employed in the passive dosing experiments (13 °C). The $K_{PDMS/w}$ of large non-polar chemicals is expected to increase with decreasing temperature. Enthalpies of phase change from PDMS to water between −21 and −46 kJ mol^{-1} have been reported for several PAHs (Muijs and Jonker, 2009). Assuming, in the absence of measured values for the chemicals studied here, an average value of −35 kJ mol^{-1}, $K_{PDMS/w}$ would increase by a factor 1.8 when the temperature decreased from 25 °C to 13 °C. This is insufficient to explain the difference reported in Table 2.

It could also be that there are large variations in $K_{PDMS/w}$ between manufacturers, and that the PDMS used in the PermSelect® unit had particularly high $K_{PDMS/w}$ values. However, in their review of available $K_{PDMS/w}$ data, DiFilippo and Eganhouse (2010) concluded that differences in $K_{PDMS/w}$ between manufacturers were insignificant. Furthermore, Smedes et al. (2009) determined $K_{PDMS/w}$ for silicones from different manufacturers and found that the values differed by 0.18–0.56 log units, which is in good agreement with the study by ter Laak et al. (2008) who reported a similar range for four PDMS sampling materials. The reported differences in $K_{PDMS/w}$ between PDMS manufacturers and materials are thus considerably less than the differences in Table 2.

Another possibility is that the mass of PDMS in the PermSelect® unit was much greater than estimated from the product data sheet (38 g). To test this we disassembled a unit and weighed the PDMS tubing, obtaining 40 g. This agreed with the original estimate, so an underestimation of the PDMS volume could be discarded as an explanation for the order of magnitude higher than expected partitioning capacity of the unit.

The most likely explanation would appear to be that other materials in the passive dosing unit besides PDMS made the major contribution to the chemical sorption capacity. As noted above, the unit also contained polycarbonate, polypropylene, and acrylic resins. This unexpectedly high sorption capacity is beneficial for this application, as it expands the range of conditions over which the unit can be employed without chemical depletion occurring.

The fact that other materials besides PDMS dominated the sorption capacity means that the performance of the passive dosing system cannot be readily predicted from just $K_{PDMS/w}$ and the volume of PDMS in the unit. However, it is possible to build a model based on the empirical observations made here. Although PDMS was not the dominant chemical reservoir in the unit, there was still a strong relationship between $K_{PDMS/w}$ and $K_{ds/w}$. A linear regression of $\log K_{ds/w}$ against $\log K_{PDMS/w}$ gave:

$$\log K_{ds/w} = 0.785 \log K_{PDMS/w} + 0.700; \qquad r^2 = 0.83 \tag{3}$$

or

$$m = 5.0 K_{PDMS/w}^{0.785} C_W \tag{4}$$

where m is the mass of the chemical (in g) loaded into the unit. Eq. (4) can be used to estimate the required loading of the passive dosing unit in order to achieve an aqueous concentration C_W (g L^{-1}) for a chemical with known $K_{PDMS/w}$. The analogous equations for K_{OW} are:

$$\log K_{ds/w} = 0.630 \log K_{OW} + 0.874; \qquad r^2 = 0.83 \tag{5}$$

and

$$m = 7.5 K_{OW}^{0.630} C_W. \tag{6}$$

They can be employed when an estimate of $K_{PDMS/w}$ is not readily available. Similarly, the half life for the aqueous concentration produced by the passive dosing unit can be estimated from:

$$t_{50} = \frac{3.5 K_{PDMS/w}^{0.785}}{q} \tag{7}$$

and

$$t_{50} = \frac{5.2 K_{OW}^{0.630}}{q} \tag{8}$$

where q is the (continuous) water flow rate through the unit (L h^{-1}) and t_{50} is the half life in h.

These models are preliminary; they can be expected to give only rough estimates of the system performance. For their further refinement it would be necessary to identify the material responsible for the higher than expected sorption capacity of the units. The quantity of this material may vary from unit to unit, which could explain some of the observed variability in unit performance with respect to both the magnitude and the half life of the aqueous concentrations. A better understanding of the exposure of this material to the loading solution might also help to improve the reproducibility of the loading procedure.

3.6. Safety aspects, applications and possibilities for improvements

In addition to producing constant aqueous concentrations of hydrophobic substances over long periods of time (i.e., weeks) and offering flexibility in dosing via regulation of the flow rate, the method requires little contact time of the operator with the chemicals. The system is closed and the only significant contact that the user has with the chemicals is in the preparation of the methanol solution for loading. Thus this method has advantages over existing alternative methods with respect to occupational hygiene.

This method can be readily applied when constant concentrations of hydrophobic chemicals are required in aqueous solutions. One benefit over several other methods is that it eliminates the risk of free droplets or grains of chemical entering the test system. It is not restricted to generating concentrations close to the aqueous solubility, as it operates according to a partitioning principle. Hence it is not dependent on a regulated dilution stream in order to establish the desired concentrations. It has several obvious applications in organic environmental chemistry, including generating constant water concentrations for bioaccumulation and toxicity experiments with aquatic macroflora and -fauna. It could also be extended to generate stable concentrations in air for conducting experiments with terrestrial organisms.

The applied hollow fibre device had not been designed with passive dosing in mind. It contained several polymeric phases, each of which presumably had a considerable partitioning capacity for the investigated chemicals. While this multi-phase device proved to be well suited for generating stable concentrations, it was not optimal with regards to accurately setting predetermined aqueous concentrations based on PDMS/water partition coefficients, which is possible with simpler passive dosing formats that are used for smaller test systems (Smith et al., 2010). The hollow fibre device could be optimised by a careful selection of materials that have very limited sorptive capacity compared to the silicone microtubes, particularly at the outflow. Such an optimisation of materials has recently been shown to significantly improve the performance of

hollow fibre modules for equilibrium sampling through membranes (Larsson et al., 2009), and a similar improvement is expected for flow-through passive dosing.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.chemosphere.2011.10.024.

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