Partitioning behaviour of isothiocyanates in the two-phase system 1-octanol/water

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The distribution kinetics of 20 isothiocyanates in two-phase 1-octanol/water system was characterized by transport rate parameters relating the flux through the interface to concentration of the solute in the aqueous and the organic compartment (l_1 and l_2, respectively). The parameters were shown to be dependent on the partition coefficient $P = l_1/l_2$ as:

$$\log (l_1/(\mathrm{m\,s^{-1}})) = \log P - \log (\beta P + 1) + \mathrm{const}$$

$$\log (l_2/(\mathrm{m\,s^{-1}})) = -\log (\beta P + 1) + \mathrm{const}$$

$$\mathrm{const} = -4.907,\ \beta = 0.261$$

Importance of this finding for assessment of distribution of the compounds in biological systems is discussed.

Кинетика распределения 20 изотиоцианатов в двухфазовой системе 1-октанол/вода была характеризована скоростными данными транспорта. Эти данные являются константой пропорциональности между течением через межфазовый раздел и концентрацией транспортированного вещества в водной и органической фазах (l_1 и l_2) и зависят от коэффициента распределения $P = l_1/l_2$:

$$\log (l_1/(\mathrm{m\,s^{-1}})) = \log P - \log (\beta P + 1) + \mathrm{const}$$

$$\log (l_2/(\mathrm{m\,s^{-1}})) = -\log (\beta P + 1) + \mathrm{const}$$

$$\mathrm{const} = -4{,}907,\ \beta = 0{,}261$$

Значение этого определения для распределения веществ в биологических системах обсуждается.

Isothiocyanates are naturally occurring or synthetic compounds with broad spectrum of antimicrobial and cytotoxic activities [1]. They enter the cell by diffusion through lipidic part of the membrane, *i.e.* by the same mechanism as most of xenobiotics [2] and also some metabolites and cell constituents (ethanol, urea, glycerol) [3]. As this process is independent of structure of both the transported solutes and membrane components [2] it can be studied advantageously in model systems [4], one of the simplest being the two-phase system

water—immiscible organic solvent [5]. Its interface physically resembles that existing between the membrane and surrounding aqueous medium due to the presence of diffusion layers on aqueous as well as on "organic" side in both the cases. This approach makes it possible to study in more detail the key step, *i.e.* the transition of the transported substance from aqueous to unpolar phase.

The kinetics of the process is characterized by transport rate parameters l_1 and l_2 (direction water—organic solvent and backwards, resp.). The kinetic parameters were shown to be dependent on the partition coefficient $P = l_1/l_2$ [6—8]

$$\log \{l_1\} = \log P - \log (\beta P + 1) + \text{const} \qquad (1)$$

$$\log \{l_2\} = -\log (\beta P + 1) + \text{const} \qquad (2)$$

where the constants β and const are dependent on the model system.

In this study distribution kinetics of 20 isothiocyanates is investigated in 1-octanol/water system with the aim to verify the published relationships (*1*) and (*2*). In addition, the dynamic method was used to determine the partition coefficients of the compounds tested as classical shake-flask method [2] is unsuitable due to instability of isothiocyanates in aqueous solutions [1].

Experimental

The experiments were performed in the all-glass apparatus described in [9]. The two-phase 1-octanol/water system was stirred with frequency 1.3 Hz satisfying that no concentration gradients could appear in either phase and the motion of the phase interface was minimal. At constant stirring rate the size of the interface was virtually constant, its shape being slightly conical.

The measuring vessel was filled with redistilled deionized 1-octanol—saturated water (175 cm^3), temperated to 25°C and surfaced with the solution of the respective compound in freshly distilled 1-octanol saturated with water (25°C, 20 cm^3). At appropriate time intervals samples (2 cm^3) for spectrophotometric determination of the solute concentration were withdrawn with a syringe. The UV and VIS spectra were recorded in the whole spectral range to monitor the stability of compounds in the given medium in addition to their concentrations.

Transport of a compound in the two-phase system can be described by a system of two linear differential equations

$$-\mathrm{d}c_1/\mathrm{d}t = (Sl_1/V_1)\, c_1 - (Sl_2/V_1)\, c_2 \qquad (3)$$

$$-\mathrm{d}c_2/\mathrm{d}t = -(Sl_1/V_2)\, c_1 + (Sl_2/V_2)\, c_2 \qquad (4)$$

where c stands for concentration of the compound, V for volume (subscripts 1 and 2 indicate aqueous and 1-octanol phases, resp.), S for surface of the

interface, l for transport rate parameters in direction water—1-octanol and backwards (subscripts 1 and 2, resp.), and t for time. Provided that the compound is present in 1-octanol only at the beginning of partition in the concentration c_0, the time course of concentration in aqueous phase (c_1) can be expressed by the equation

$$c_1 = c_0 l_2 V_2 [1 - \exp(-S(l_1/V_1 + l_2/V_2)\, t)]/(l_1 V_2 + l_2 V_1) \qquad (5)$$

As for the time course of absorbance in the aqueous phase, it is described by eqn (*5*) multiplied by molar absorption coefficient ε and the length of light path d.

The values of transport rate parameters l_1 and l_2 were determined by nonlinear regression analysis [10] according to eqn (*5*) expressed through absorbance, the initial estimates of l_1 and l_2 being made on the basis of eqns (*6*) and (*7*) resulting from eqn (*5*)

$$\ln(A_e - A) = \text{const} - S(l_1/V_1 + l_2/V_2)\, t \qquad (6)$$

$$A = \varepsilon d c_0 l_2 S t / V_1 \qquad (\text{for } t \to 0) \qquad (7)$$

where A is absorbance, subscript e indicates attainment of distribution equilibrium, *i.e.* cancelling of the time term from eqn (*5*). Eqn (*7*) represents eqn (*5*) expressed through absorbance with its exponential part being substituted by the first two terms of the corresponding Taylor series for time approaching zero.

Results and discussion

Structures, transport rate parameters l_1 and l_2 as well as the partition coefficients P of the derivatives tested are summarized in Table 1. As indicated by the values of statistical parameters, eqn (*5*) is a good description of distribution kinetics. Consequently, hydrolysis of the compounds [1] under the given conditions is negligible. Otherwise it would be necessary to take this process into account and modify the corresponding eqns (*3*—*7*).

The dependence of transport rate parameters l_1 and l_2 on the partition coefficient P can be described by eqns (*1*) and (*2*) with the following values of adjustable parameters optimalized by nonlinear regression analysis: const $= -4.907$, $\beta = 0.261$, statistical parameters being number of points $n = 40$, correlation coefficient $r = 0.996$, standard deviation $s = 0.162$ and F-test $F = 4390$. The values of const and β are in good agreement with those published previously for 2-furylethylenes and determined under identical experimental conditions [9] (in that paper the value of const $= -5.600$ is erroneous and ought to be replaced by -4.976, $\beta = 0.261$). The difference between ours and the *van de Waterbeemd*'s values of const and β [7, 8] is caused probably by

various quality of diffusion layers on the interface resulting from different hydrodynamics of both test systems.

Table 1

Structures of the investigated isothiocyanates (R-NCS), transport rate parameters in direction water—1-octanol (l_1) and backwards (l_2), the partition coefficients $P = l_1/l_2$. The values of l_1 and l_2 were determined by nonlinear regression analysis [10] of kinetic data according to eqn (5). In individual experiments, the lowest values of number of experimental points, correlation coefficient, and F-test were $n = 11$, $r = 0.987$, and $F = 111$, the maximum value of standard deviation $s = 0.038$

R	$-\log(l_1/(\mathrm{m\,s^{-1}}))$	$-\log(l_2/(\mathrm{m\,s^{-1}}))$	$\log P$
CH_3	4.727	5.665	0.938
$CH_3(CH_2)_3$	4.337	7.255	2.918
$CH_3CH(CH_3)CH_2$	4.555	7.379	2.824
$CH_3CH_2CH(CH_3)$	4.251	7.064	2.813
C_6H_5	4.158	7.527	3.369
4-CH_3—C_6H_4	4.198	8.100	3.902
3-CH_3O—C_6H_4	4.291	7.970	3.679
4-CH_3O—C_6H_4	4.230	7.809	3.579
3-COOH—C_6H_4	4.508	6.909	2.401
4-COOH—C_6H_4	4.489	6.929	2.440
3-CN—C_6H_4	4.289	7.418	3.129
4-CN—C_6H_4	4.319	6.973	2.654
4-$N(CH_3)_2$—C_6H_4	4.283	8.242	3.959
3-OH—C_6H_4	4.290	7.524	3.234
3-Cl—C_6H_4	4.141	7.502	3.361
4-Cl—C_6H_4	4.241	8.147	3.906
3-Br—C_6H_4	4.261	8.378	4.117
4-Br—C_6H_4	3.885	7.868	3.983
4-I—C_6H_4	4.078	8.296	4.218
4-NO_2—C_6H_4	4.262	7.809	3.547

It can be concluded that the distribution kinetics of low-molecular-mass compounds in a two-phase system is, in fact, structurally independent and determined by the only physicochemical property — the partition coefficient. Quality of the interface is involved in the terms const and β. Van de Waterbeemd showed that const is decadic logarithm of the rate constant for transport through organic diffusion layer and β is the ratio of rate constants characterizing diffusion through organic and aqueous diffusion layers.

In view of solute transport, biological systems can be considered as composed of alternating aqueous and lipidic phases. The results presented can be used for estimation of concentration in individual compartments of biological systems [11—14].

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