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Does the size of small objects influence chemical reactivity in living systems?

La taille des petits objets a-t-elle une influence sur la réactivité chimique dans les systèmes vivants ?

Albert Sanfeld [*], Albert Steinchen

'Thermodynamique–Systèmes chimiques complexes', UMR 6171, faculté des sciences de Saint-Jérôme, bd Escadrille-Normandie-Niemen, 13397 Marseille cedex 20, France

Presented by Jacques Ricard

Abstract

Previous theoretical works showed that chemical reactions in micro- and nano-droplets, bubbles and solid particles were strongly affected by their confinement. In particular, the smallness of the systems leads to high internal pressure compared to the external pressure, which then significantly modifies the values of chemical equilibrium and kinetic constants. In addition, surface tension or surface stress, reactional dilatation and surface charge play also a major role on the chemical reactivity. As living systems are also made of very complex dispersed subsystems, i.e. organelles, it seemed obvious to illustrate our theory by some biological actual examples encountered in pulmonary alveolæ, in vacuolæ and in medical applications, such as dissolution of gallstones. *To cite this article: A. Sanfeld, A. Steinchen, C. R. Biologies 326 (2003).*
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Résumé

Nos études théoriques précédentes ont montré l'influence de la taille de micro- et nano-gouttes, bulles et particules solides sur la réactivité chimique et le transfert de matière. La complexité du milieu réactif est liée ici à l'état multiphasique, comprenant de très petits objets (organelles), et non à la réactivité intrinsèque. En vertu de la loi de Laplace, la petitesse de ces objets leur confère un état d'extrême pression, modifiant les constantes chimiques et cinétiques. Une telle situation est le résultat des effets de tension superficielle, de dilatation réactionnelle et de la charge électrique de l'objet, qui modifient considérablement les valeurs des constantes. Nous avons alors tout naturellement étendu l'objet de nos investigations aux réactions se déroulant au sein d'organelles dans les systèmes vivants. Nous avons notamment appliqué notre théorie au calcul de constantes d'équilibre de réactions dans l'alvéole pulmonaire, à la réaction de dissolution de calculs biliaires et au calcul de la constante cinétique

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[*] Corresponding author.
E-mail address: a.sanfeld@vmesa12.u-3mrs.fr (A. Sanfeld).

1. Introduction

It is well known that complexity in biology results in kinetics of numerous mechanisms, which involve high intrinsic non-linearities. For example, enzymatic reactions far from equilibrium may lead to steady and unsteady unstable states with, however, either local organization such as space structures, limit cycles and multistates transitions or deterministic chaos [1,2]. The same situation may also arise when more simple kinetics take place in particular conditions: presence of curved interfaces [3], effects in electric fields [4], transition critical point [5]. The coupling with hydrodynamic processes in models of fluid biological membranes was also taken into account [6]. On the basis of the bifurcation theory, the fundamental background of all these fascinating phenomena was formulated and developed by the Brussels school of Prigogine, Glansdorff, Nicolis and co-workers at the end of the last century [7,8]. They enter the frame of the so-called order by fluctuations, which creates local dissipative structures in far-from-equilibrium conditions.

Another kind of unexpected situations, however, not resulting from the non-linear character of the chemical mechanisms, may arise from the complexity of the space environment in which the chemical reactivity takes place. In other words, in addition to the intrinsic role of the chemical reactivity, we have now to analyse the geometric aspect of the domain concerned and the nature of the boundary conditions. This justifies the interest in the study of reactions and matter transfers in strong confinement encountered in disperse media containing micro (1–10 µm) and nano (10^{-2}–10^{-3} µm) objects, droplets, bubbles, solid crystals or particles. All these small systems are, nevertheless, still considered as macroscopic entities, according to the magnitude of the fluctuations. They behave as chemical reactors submitted to very high internal pressures (10^2–10^4 atm) compared to the external pressure. Hence, we then expect that thermo-dynamic and kinetic properties should be significantly influenced by the smallness of the size of the object. This was shown in detail in our previous papers through numerous examples from atmosphere, sonochemistry and metallurgy [9,10]. One of the main interests, among other points, is that small objects give the potentiality to obtain products of reactions that would not be formed in large reservoirs. We will summarize in the next two sections the main theoretical results obtained for the influence of the confinement on some thermodynamic and kinetic properties, and then put the emphasis on applications to living systems in the last section.

2. Influence of the size of an object on the equilibrium constant

Among the thermodynamic and kinetic properties we shall restrict our final result to the chemical equilibrium and kinetic constants, by comparing the same reaction at the same temperature and the same external pressure, for small and large non-deformed objects. We first start with uncharged systems.

2.1. Uncharged systems

As well known from classical thermodynamics, the chemical equilibrium constant K is defined by:

$$K = \exp \frac{A^0}{\mathcal{R}T} \qquad (1)$$

where the standard chemical affinity A^0 $(= -\Delta G^0)$ is written in terms of the standard chemical potentials μ_i^0 of the species i involved in the reaction:

$$A^0 = - \sum_i \nu_i \mu_i^0 (T, p) \qquad (2)$$

with ν_i the stoichiometric coefficient. We must recall here that the stoichiometric composition ratio is equal to the equilibrium constant only for compositions at

equilibrium. Hence, the theory presented here is not restricted to thermodynamic equilibrium states.

In a liquid or in a solid phase, the standard chemical potential reads:

$$\mu_i^0 = \mu_i^{00}(T) + p v_i^0 \left(1 - \chi_i \frac{p}{2}\right) \tag{3}$$

where v_i^0 is the standard molal volume extrapolated at zero pressure, which depends only on T, and χ_i the compressibility coefficient of i in the mixture; the term $\chi_i \frac{p}{2}$ is often negligible ($\ll 1$), except for microdroplets of liquid metals.

In a vapour phase, the standard chemical potential reads

$$\mu_i^0 = \mu_i^{00}(T) + \mathcal{R}T \ln p \tag{4}$$

At the surface of tension of a spherical object and under mechanical equilibrium conditions, the classical Laplace law may be applied:

$$\Delta p = \frac{2\sigma}{R} \quad \text{(droplets and bubbles)} \tag{5}$$

where Δp is the jump of pressure between the internal phase of the object and the external continuous phase, σ is the surface tension and R the radius of the spherical object. For large objects ($R \to \infty$), however, the internal and external pressures are equal in the absence of external forces. At the surface of tension of an isotropic solid crystal or amorphous solid, the jump of pressure Δp is related to the change of the standard chemical potential and reads:

$$\Delta p = \frac{2S}{R} \tag{6}$$

where S is the surface stress and $2S/R$ the capillary-elastic pressure (for more details on this very complicate and controversial question, see [10,11]). By comparing the same reaction at the same temperature and external pressure, for small and large objects (superscript*), the ratio of the chemical equilibrium constant becomes respectively:

$$\frac{K}{K^*} = \exp\left\{\left[-2\sigma^*/R + 4\sigma^* \Gamma_s v_s^{0l}/R^2\right]\Delta^{0l}\right\}$$
spherical droplet $\tag{7}$

$$\frac{K}{K^*} = \left\{1 + \frac{2\sigma^*}{p^*R}\left(1 + 2\Gamma_s v_s^{0l}/R\right)\right\}^{-\nu}$$
spherical bubble $\tag{8}$

$$\frac{K}{K^*} = \left\{1 + \frac{2}{p^*R}\left(\sigma + 2\eta \, dR/dt\right)\right\}^{-\nu}$$
expanding or shrinking bubble $\tag{9}$

$$\frac{K}{K^*} = \exp\left\{[-2S/R]\Delta^0\right\}$$
spherical solid particle $\tag{10}$

$$\frac{K}{K^*} = \exp\left\{[-4S/l]\Delta^0\right\} \quad \text{cubic crystal} \tag{11}$$

where Γ_s and v_s^0 are the surface concentration and the standard molal volume of the solvent, Δ^0 the reactional dilatation and ν the global stoichiometric coefficient respectively defined by:

$$\Delta^0 = \sum_i \nu_i v_i^0/\mathcal{R}T \tag{12}$$

$$\nu = \sum_i \nu_i \tag{13}$$

and l is the size of the cube, η is the bulk shear viscosity of the continuous phase, σ^* is the surface tension of the same interface for an infinite radius of curvature. The second member of the exponent in equations (7) and (8) accounts for the influence of the curvature on the surface tension. The quantity $\eta\frac{dR}{dt}$ in equation (9) is the resistance opposed by the surrounding fluid to the expansion or shrinkage of the bubble.

Remark. When dealing with chemical reactions (including matter transfers) in multiphase systems, the ratio K/K^* appears only through the reactive components inside the confined region.

2.2. Charged systems

When a reaction occurs in a spherical charged droplet in the presence of a vapour phase, the induced electric field polarizes the vapour, the hydrostatic pressure and the surface tension being then dependent on the intensity of the field [9,12]. The ratio K/K^* reads then:

$$\frac{K}{K^*} = \exp\left\{\left[-2\sigma^*/R + 4\sigma^* \Gamma_s v_s^{0l}/R^2 \right.\right.$$
$$\left.\left. + Q^2/8\pi R^4\right]\Delta^{0l}\right\} \tag{14}$$

where Q is the global charge of the droplet. Consider now a reaction that involves j components in the

vapour phase surrounding a charged droplet, we get:

$$\frac{K}{K^*} = \left(1 + \frac{Q^2}{8\pi p^* R^4}\right)^{-\nu}$$

$$\times \exp\left\{\frac{Q^2\left(\sum_j \alpha_j + \frac{m_j^2}{3kT}\right)}{2kTR^4}\right\} \tag{15}$$

where α_j is the molecular polarizability, $\underline{m}_j$ the permanent dipole moment of component j, and $\nu = \sum_j \nu_j$. The factor of the exponential refers to the influence of the electric field on pressure, while the exponent refers to the polarization of the matter in the vapour.

The predictions to be obtained from equations (7)–(11), (14) and (15) are the following:

- (*i*) high values of surface tension or of surface stress and high curvature influence the equilibrium constant, the effect being, however, much more important for bubbles than for droplets, and also important for small solid particles according to their high surface stress;
- (*ii*) charges on droplets reduce this effect (equation (14));
- (*iii*) the equilibrium constant of reaction taking place in the vapour in contact with a charged droplet depends on the electric field pressure and weakly on the polarization term (equation (15));
- (*iv*) the expansion (or shrinkage) contribution ($\eta\, dR/dt$) plays a significant role only in very viscous media (equation (9));
- (*v*) the role of the global stoichiometry of the reaction is important for reactions in bubbles (equations (8) and (9)) or in vapour around droplets (equation (15)) as well as the reactional dilatation in droplets and solid particles (equations (7), (10), (11) and (14)).

Remark. For a charged droplet in the presence of an insoluble liquid phase, we must take into account the thickness of the double layer established in the presence of ionic amphiphilic molecules. The prediction on K/K^* should be qualitatively identical to those obtained for the droplet in presence of vapour.

3. Influence of the size of a bubble on the kinetic constant

In the Eyring–Polanyi theory of the activated complex, it is assumed that the activated complex X is in equilibrium with the initial components.

A reaction $\nu_1\,M_1 + \nu_2\,M_2 \rightleftharpoons \nu_3\,M_3 + \nu_4\,M_4$ between molecules M may be decomposed in two reactions involving the formation of an activated complex: a direct reaction of formation of the complex $\nu_1\,M_1 + \nu_2\,M_2 \xrightarrow{\vec{k}_a} X \rightarrow \nu_3\,M_3 + \nu_4\,M_4$ and the reversed reaction $\nu_1\,M_1 + \nu_2\,M_2 \xleftarrow{\overleftarrow{k}_a} X \leftarrow \nu_3\,M_3 + \nu_4\,M_4$ where $\vec{k}_a$ and $\overleftarrow{k}_a$ are the direct and antagonist kinetic constants related to the standard affinities of $\vec{A}_X^0$ and $\overleftarrow{A}_X^0$. The kinetic constants read then:

$$\vec{k}_a = \frac{kT}{h}\frac{1}{f_X v}\left\{\exp\frac{\vec{A}_X^0}{\mathcal{R}T}\right\} \tag{16}$$

$$\overleftarrow{k}_a = \frac{kT}{h}\frac{1}{f_X v}\left\{\exp\frac{\overleftarrow{A}_X^0}{\mathcal{R}T}\right\} \tag{17}$$

where kT/h is the frequency factor, f_X, the activity coefficient of the activated complex and v is the mean molar volume.

Comparing, for the same reaction of activation, at the same temperature and external pressure, the direct kinetic constant in a small bubble and in a non confined gas, we get from equations (2), (4), (5) and (16):

$$\frac{\vec{k}_a}{\vec{k}_a^*} = \frac{f_X^* v^*}{f_X v}\left\{\frac{1}{1 + \frac{2\sigma}{Rp^*}}\right\}^{(|\nu_1+\nu_2|)-1} \tag{18}$$

In the approximation of perfect gas, by using again Laplace law, equation (18) reads:

$$\frac{\vec{k}_a}{\vec{k}_a^*} = \left\{1 + \frac{2\sigma}{Rp^*}\right\}^{(|\nu_1+\nu_2|)} \tag{19}$$

For a pair collision ($\nu_1 + \nu_2 = +2$) hence, equation (19) becomes finally:

$$\frac{\vec{k}_a}{\vec{k}_a^*} = \left\{1 + \frac{2\sigma}{Rp^*}\right\}^2 \tag{20}$$

4. Reactions in living system

4.1. Influence of the size of a pulmonary alveola on typical reactions

A pulmonary alveola is formed by one layer of epithelial cells separated from the gas by a layer of 1 μm, called hypophase (Fig. 1). Phosphoglyceric components and particularly lecithins are adsorbed at the interphase air–hypophase. In practice, for human newborn children and adults, the typical radii lie between 5 and 70 μm. As lecithin is a surfactant, during compression–depression cycle of the interfacial film [13,14], the surface tension varies from a very weak value close to 0 to 50 mJ m^{-2} (Fig. 2).

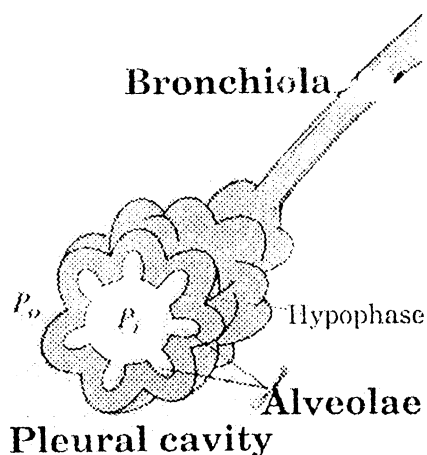Decomposition of the nickel carbonyl accidentally present in the atmosphere may occur in the pulmonary
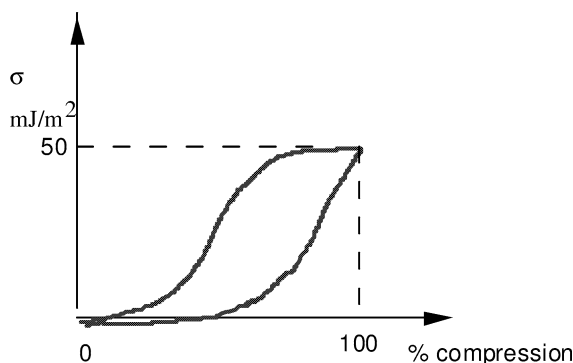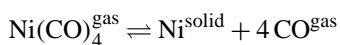

Fig. 1. Pulmonary alveola.

Fig. 2. Surface tension in compression cycle.

Table 1
Ratio of equilibrium constant versus radius of alveolæ for nickel carbonyl decomposition

R (μm)	K/K^*
5	0.58
10	0.75
50	0.94
100	0.96

alveolæ following the stoichiometric scheme:

$$\text{Ni(CO)}_4^{\text{gas}} \rightleftharpoons \text{Ni}^{\text{solid}} + 4\,\text{CO}^{\text{gas}}$$

Considering as a first approximation that the alveola is a sphere, we calculate K/K^* from equation (8). Taking $\sigma = 50$ mJ m^{-2} and $p^* = 1$ atm, we obtain K/K^* for different values of the radius R (Table 1).

Hence, the smallest the size of the alveola, the less efficient is the nickel carbonyl decomposition.

4.2. Role of the size of a solid dust in pulmonary alveola

Let us now draw our attention to the influence of the size of Ni particle on the equilibrium constant of the Ni carbonyl decomposition. Due to the breaking of the lattice of Ni, which creates vacancies and thus an increase of atomic volume, we may reasonably assume that $v_{\text{Ni}}^0 \cong 13$ cm^3 mol^{-1}. In a vacuum, $S_{\text{Ni}} \cong 2500$ mJ m^{-2}. However, in the alveola, Ni is oxidised, so that $S_{\text{Ni}}^{\text{oxidised}} \cong 1500$ mJ m^{-2}. Due to the interactions with proteins present in the alveolar hypophase, the surface stress of Ni may be considerably lowered and $S_{\text{Ni}}^{\text{coated}} \cong 130$ mJ m^{-2}. In the presence, however, of proteases that destroy the proteins, one could expect that $500 < S_{\text{Ni}}^{\text{coated+protease}} < 800$ mJ m^{-2}. Let us chose $S_{\text{Ni}} \cong 600$ mJ m^{-2} and $T = 310$ K. Under these conditions, we obtain the values of K/K^* displayed in Table 2 by using equation (10).

Table 2
Influence of the size of a Ni particle on the equilibrium constant of decomposition of Ni carbonyl in the lung alveola
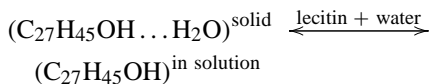
R (μm)	K/K^*
1	0.99
0.1	0.94
0.01	0.54
0.005	0.30

In conclusion, the presence of large Ni particles in the pulmonary alveolæ favours the decomposition of Ni carbonyl.

4.3. Gallstones dissolution

As shown by Houton et al. [15], cholesterol particles are solved in a mixture water–lecithin and form micelles.

The reaction reads:

$$(C_{27}H_{45}OH\ldots H_2O)^{solid} \xleftarrow{\text{lecitin + water}}$$

$$(C_{27}H_{45}OH)^{in\ solution}$$

For a spread monolayer of cholesterol on water, the surface tension equals 22 $mJ\,m^{-2}$ [16] at room temperature. We may reasonably assume that for hydrated cholesterol crystal, S should be of the order of 35 $mJ\,m^{-2}$. To give an estimate of the role of the size of the gallstones (in the presence of water $+$ lecithin) on the dissolution equilibrium constant, we choose the values [17]: $S = 25$ $mJ\,m^{-2}$, $v_{chol}^0 = 231.5$ $cm^3\,mol^{-1}$ at $T = 300$ K. The values obtained from equation (11) are summarized in Table 3.

As expected, small crystals are more soluble than large ones.

Table 3
Role of the size of a cubic crystal of gallstones on the dissolution equilibrium constant

l (µm)	K/K^*
1	1
0.1	1.1
0.01	2.5
0.005	6.4

4.4. Role of the size of a gas vacuola on the kinetic constant

Consider a binary elementary collision leading to an activated complex. The reaction of formation of H_2S in a vacuola of a sulphur bacteria (Fig. 3) [18] might proceed by such a mechanism.

For a reasonable value of $\sigma = 30$ $mJ\,m^{-2}$ and $p^* = 1$ atm, values of $\vec{k}_a/\vec{k}_a^*$ are obtained from equation (20) for any kind of binary activation process in a perfect gas phase (Table 4).

The smallness of the alveola has thus an important influence on the kinetic constant. In agreement with
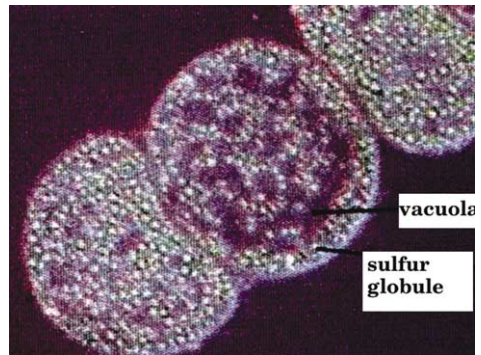

Fig. 3. Vacuolæ and sulphur globules in Thiomargarita (from [18]).

Table 4
Role of the size of a gas vacuola on the kinetic constant for any binary activation process

R (µm)	$\vec{k}_a/\vec{k}_a^*$
10	1.1
5	1.25
1	2.6
0.5	4.9
0.1	50

the Eyring's theory, high confinement increases significantly the activation process.

5. Conclusion

Whether the biologist is concerned with the properties of a system, with an equilibrium constant, or with a reaction rate, the solution of the problem requires not only a knowledge of the intrinsic mechanisms but also of the confinement in which they take place. To understand such a problem, one must know the nature of the space environment, which, in living systems, is usually a very complex region. In particular, the existence of micro- and nano-subsystems like organelles in which numerous biochemical reactions occur, may significantly influence thermodynamic and kinetic properties. One must also know fundamental relations that come under statistical mechanics, electrostatics and capillarity. As it is not yet possible to carry out such a programme in every detail, as already presented in previous papers [10,11], we have restricted our goal to give in a concise form a brief comprehensive survey of the equations we need. They clearly show the in-

fluence of the size of objects (droplets, bubbles, solid particles) and of surface tension (or surface stress) on equilibrium and kinetic constants. To illustrate the interest of our theoretical approach, we have presented examples of biological interest: reaction in pulmonary alveolæ, dissolution of gallstones and reactions in gas vacuolæ of bacteria. It appears that the confinement may significantly influence chemical constants. Other examples in living systems are at present under investigation and shall be submitted to publication later on. According to our results, attention must also be drawn on experiments performed in tubes as the replicas of real processes that occur in living systems. In addition, artefacts may be also induced by external effects. For example, by sonication and depending on the frequencies imposed, cavities are created, which modify the chemical reactivity, as shown in our previous paper [9].

Acknowledgements

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