TRANSPORT OF CARBON DIOXIDE ACROSS A MICROBIAL FILM AT THE GAS–WATER INTERFACE

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During the transport of CO_2 across a film of microbial suspension the gradient of CO_2 in the film is affected by the hydration of CO_2 and the actual activity of microorganisms producing or consuming CO_2. If the pH gradient in the film along the diffusion path is negligible one may derive expressions for the mass flow of CO_2 across the suspension–gas interface. For the limiting case of CO_2 transport in the cultivation medium (without pronounced microbial activity as to CO_2) these expressions are compared with experimental data on CO_2 absorption in a phosphate buffer and in a cultivation medium for algae. A qualitative agreement between the theoretical relationship and experimental data is observed.

During cultivation of microorganisms carbon dioxide may be consumed (*e.g.* by autotrophic algae) or given off (*e.g.* aerobic microorganisms). It is essential for a satisfactory cultivation that the level of CO_2 in the microbial suspension be kept below a critical value so as to influence unfavourably the metabolic processes taking place in the cells.

The cultivation liquid usually contains H_2CO_3, acid groups of proteins, phosphoric acid and organic acids which all act as H^+ donors; HCO_3^-, negatively charged protein groups, HPO_4^{2-} of organic phosphates, these all acting as H^+ acceptors (*e.g.* Nyiri and Lengyel[1]). During the cultivation of microorganisms, the buffering capacity of the external medium may change and this affects the rate of hydration and dehydration of CO_2. At low and medium values of pH at which microorganisms are usually cultivated, the cultivation liquid contains both free CO_2 and HCO_3^- ions, the question of ability of microorganisms to utilize this anion being still unresolved.

Concentration of CO_2 within the suspension is affected by CO_2 transport across the interface between suspension and the gaseous phase, the main resistance to transport being concentrated in the suspension film. The concentration gradient of CO_2 in the film may be affected by the reversible hydration reaction which may be catalyzed by some components of the cultivation medium[1,2] such as phosphate ions. With more rapidly growing microorganisms one must take into account the produced or utilized CO_2 and its effect on CO_2 transport across the suspension film. The region of CO_2 transport between gas and liquid at roughly neutral pH when most biosynthetic processes take place has not been sufficiently explored although it is of utmost importance for microbial cultivation of various types.

THEORETICAL

Let us consider a suspension film across which CO_2 diffuses from the suspension to the gaseous phase (desorption) or from the gaseous phase into the suspension (absorption). The balance of CO_2 in a steady state is given by:

$$D(\mathrm{d}^2C/\mathrm{d}x^2) - r \pm R = 0\,, \tag{1}$$

where D is the diffusivity of CO_2 in the suspension, C the concentration of free CO_2, x the distance from the interface, r the rate of CO_2 consumption by hydration reaction $\pm R$ the rate of formation (+) or consumption (−) of CO_2 by microorganisms. The hydration of CO_2 proceeds according to

$$CO_2 + H_2O \overset{k}{\rightleftharpoons} HCO_3^- + H^+\,. \tag{A}$$

The rate constant k of this reaction generally depends on the catalytic effect of cultivation medium components. The rate of CO_2 consumption is thus

$$r = k(C - C_e) \tag{2}$$

the equilibrium concentration C_e being given by

$$C_e = BH/K_1\,. \tag{3}$$

In this expression, B and H represent the local concentrations of HCO_3^- and H^+ in the film, K_1 is the equilibrium constant of reaction (A). Generally, the values of B and H may change along the diffusion path of CO_2 in the film. In the case that the change of concentration of H^+ due to the hydration reaction of CO_2 is buffered by the cultivation medium components (donors and acceptors of H^+) one may consider the pH in the suspension film as constant and equal to the pH within the suspension. During transport of CO_2 through the suspension film one must consider the fact that CO_2 is transported also in the form of HCO_3^- ions. The balance of HCO_3^- in the film at steady state is given by:

$$D_B(\mathrm{d}^2B/\mathrm{d}x^2) + r = 0\,, \tag{4}$$

where D_B is the diffusivity of HCO_3^- in the suspension. Relationship (*4*) holds on the assumption that microorganisms do not produce (and do not take up) HCO_3^- directly. If the pH in the film is constant, equations (*1*), (*3*) and (*4*) yield:

$$\mathrm{d}^2(C - C_e)/\mathrm{d}x^2 - K(C - C_e) \pm R/D = 0\,, \tag{5}$$

where K is defined by

$$K = k[(1/D) + (H/K_1 D_B)]\,. \tag{6}$$

Let us now consider a case when the microorganisms produce CO_2 which passes from the centre of the suspension to the gaseous phase. This may be the case of ventilation of CO_2 from a culture of aerobic microorganisms (bacteria, yeasts, *etc.*). For a constant value of R, Eq. (*5*) will yield a general solution for our case:

$$\Delta C = a_1 \exp(x\sqrt{K}) + a_2 \exp(-x\sqrt{K}) + R/KD\,, \tag{7}$$

where $\Delta C = (C - C_e)$; a_1 and a_2 are constants.

For the following limiting conditions

$$x = 0\,, \quad \mathrm{d}(\Delta C)/\mathrm{d}x = \mathrm{d}C/\mathrm{d}x$$
$$x = \delta\,, \quad \Delta C = (\Delta C)_\delta \tag{8}$$

a_1 and a_2 are defined by

$$a_1 = a_2 + N_0/D\sqrt{K} \tag{9}$$

$$a_2 = [(\Delta C)_\delta - R/KD - (N_0/D\sqrt{K})\exp(\delta\sqrt{K})]\,.$$
$$.\,[\exp(\delta\sqrt{K}) + \exp(-\delta\sqrt{K})]^{-1}\,. \tag{10}$$

In this expression, N_0 stands for the mass flow of CO_2 across the interface, δ is the thickness of the film.

Equations (*2*), (*7*) and (*1*) yield a differential equation for C reflecting the case of CO_2 production by microorganisms:

$$D(\mathrm{d}^2C/\mathrm{d}x^2) - k[a_1 \exp(x\sqrt{K}) + a_2 \exp(-x\sqrt{K}) + R/KD] + R = 0\,. \tag{11}$$

Integration of this equation yields:

$$D(\mathrm{d}C/\mathrm{d}x) - k[a_1 \exp(x\sqrt{K}) - a_2 \exp(-x\sqrt{K})] :$$
$$:\sqrt{K} + R[1 - (k/KD)]\,x + a_3 = 0\,. \tag{12}$$

The integration constant a_3 is derived from equations (*9*) and (*12*):

$$a_3 = N_0[(k/KD) - 1]\,, \qquad (13)$$

where the mass flow N_0 of CO_2 is defined by:

$$N_0 = D\,\mathrm{d}C/\mathrm{d}x_{x=0}\,. \qquad (14)$$

For the limiting conditions:

$$x = 0\,, \quad C = C^+ \qquad (15a)$$

$$x = \delta\,, \quad C = C_\delta \qquad (15b)$$

integration of Eq. (*12*) and rearrangement yields:

$$K_L^+(C_\delta - C^+) - k[a_1 \exp(\delta\sqrt{K}) + a_2 \exp(-\delta\sqrt{K}) - a_1 - a_2]/K\delta + \\ + \tfrac{1}{2}R\delta[1 - (k/KD)] + a_3 = 0\,, \qquad (16)$$

where $K_L^+ = D/\delta$. Substitution for a_1, a_2, a_3 from Eq. (*9*), (*10*) and (*13*) into Eq. (*16*) and expression of K with the aid of Eq. (*6*), with suitable rearrangement, yields the final expression for CO_2 flow by desorption:

$$N_0 = \{N_0^+\gamma + K_L^+[(\Delta C)_\delta - (R/k\gamma)]\,[\cosh^{-1}(\beta\sqrt{\gamma}) - 1] + RD/2K_L^+)\}\,. \\ .\,\{\alpha + [\mathrm{tgh}(\beta\sqrt{\gamma})]\,[\beta\sqrt{\gamma}]^{-1}\}^{-1}\,, \qquad (17)$$

where $N_0^+ = K_L^+(C_\delta - C^+)$ is the flow of CO_2 by physical desorption, $\beta = (kD)^{0.5}/K_L^+$ $\gamma = 1 + \alpha$; $\alpha = HD/K_1D_B$.

In the case that CO_2 is absorbed by the microbial suspension (*e.g.*, with autotrophic algae which require CO_2 for growth) one may proceed analogously from Eq. (*1*), derive an expression analogous to (*17*) with the difference that $R/k\gamma$ is positive and $N_0^+ = K_L^+(C^+ - C_\delta)$. In the case that CO_2 is desorbed from the microbial suspension and taken up by the microorganisms (*e.g.* desorption of CO_2 from the suspension of algae cultivated on an open-air cultivation surface) the mass flow of CO_2 may be defined by an expression analogous to (*17*) with the difference that $R/k\gamma$ is positive and $RD/2K_L^+$ is negative. For the case that a chemical equilibrium exists inside the liquid as to CO_2 hydration, *i.e.* $(\Delta C)_\delta = 0$ and no microorganisms are present (or if their effect on the gradient of CO_2 in the film is negligible) Eq. (*17*) reduces to:

$$N_0/N_0^+ = \gamma[\alpha + (\beta\sqrt{\gamma})^{-1}\,.\,\mathrm{tgh}(\beta\sqrt{\gamma})]^{-1}\,. \qquad (18)$$

This is the expression for transport of CO_2 across the film in the cultivation medium. For $\alpha \ll 1$ (*i.e.* high pH values), relationship (*18*) reduces to the well-known expression defined by Hatta's number[3]:

$$N_0/N_0^+ = \beta/(\mathrm{tgh}\,\beta) = Ha\,. \tag{19}$$

Eq. (*19*) can be derived from (*1*) and (*2*) under the same assumptions as with Eq. (*18*) but with the additional assumption that the concentration of HCO_3^- in the film does not change and that it is the same as inside the liquid (C_e is constant). It is a well-known fact that with increasing pH of the liquid, the concentration ratio HCO_3^-/CO_2 will increase and one may then expect that with a sufficiently high pH of the culture the concentration of HCO_3^- is such as to make the gradient of HCO_3^- in the film negligible. Due to the buffering capacity of the cultivation medium and of microbial cells the retention of HCO_3^- in the medium will be much greater than for the noncatalyzed hydration of CO_2 in water. During transport of CO_2 in microbial systems we are generally dealing with lower concentrations of CO_2 (up to 5% by volume in the gaseous phase). With higher values of HCO_3^- retention in the system the concentration of bicarbonate ions in the film will not be much affected by CO_2 transport.

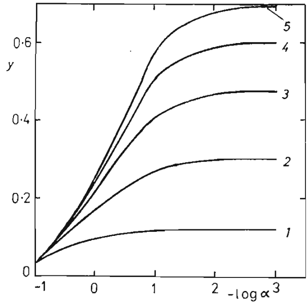

FIG. 1

Dependence of $y = \log (N_0/N_0^+)$ on $\log \alpha$ according to Eq. (*18*), the Parameter Being β

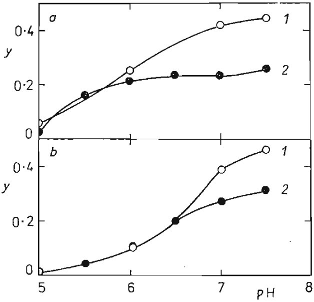

FIG. 2

Experimental Data[4] on CO_2 Absorption in 50 mM Phosphate Buffer (*a*) and in the Nutrient Medium for Algae (*b*)

Bubbled layer, liquid volume 210 ml, 20°C, rate of gas flow referred to layer cross section 4·9 cm s^{-1}, flow of liquid 0·745 (*1*) and 1·49 (*2*) litres $cm^{-2}\,h^{-1}$. $y = \log N_0/N_0^+$.

At lower pH values the CO_2 is present mainly in the free form and ratio of HCO_3^-/CO_2 concentrations decreases with decreasing pH. The assumption of a constant value of HCO_3^- concentration is then no longer justified and the limiting expression (*19*) is no more valid. Expression (*18*) must then be used. For sufficiently low values of pH then $\alpha \gg 1$ and equation (*19*) is simplified to $N_0/N_0^+ = 1$, *i.e.* the transport of CO_2 across the film is exclusively the question of diffusion of CO_2 without interaction with the hydration reaction.

Fig. 1 shows the dependence of Eq. (*18*) on dimensionless parameters α and β (parameter γ is a function of α as follows from the text under Eq. (*17*)). Figs 2*a* and 2*b* show the dependence of $\log(N_0/N_0^+)$ on the pH of the liquid derived from the experimental data of CO_2 absorption in the phosphate buffer and in the nutrient medium for algae[4] in the bubbled layer. Since $-\log \alpha$ is proportional to pH one may expect the curve obtained from the theoretical relationship (*18*) to be similar to those obtained experimentally. Fig. 1 indicates a decrease of the N_0/N_0^+ ratio as parameter β decreases, *i.e.* as K_L^+ increases. Curves 2 in Figs 2*a* and 2*b* correspond to higher values of K_L^+ than do curves *1*, *i.e.* there is an agreement with the predicted behaviour. Comparison of Fig. 1 with Figs 2*a* and 2*b* suggests a certain similarity of the trends displayed by theoretical and by experimental curves. Quantitative data on the values of α and β in microbial systems are lacking as it is difficult to determine then precisely.

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