# **Evaluation and Modeling of the Aerobic Stirred Bioreactor Performances for Fungus Broths**

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The mixing time and oxygen mass transfer coefficient represent two of the most important quantities implied on the design and operation of the bioreactors. In this paper, the influences of the main characteristics of bioreactor and of fungus broths (*P. chrysogenum*, free mycelia and mycelial aggregates) on mixing efficiency and oxygen mass transfer, are studied. By means of the experimental data, some mathematical correlations describing the influences of the considered factors on mixing time and  $k_1a$ , have been proposed. The proposed correlations can be used in bioreactor performance evaluation, optimization, and scaling-up.

Key words:

Stirred bioreactor, mixing time, mass transfer coefficient, *Penicillium chrysogenum*, pellets, free mycelia.

## Introduction

The analysis and the evaluation of bioreactor performance must respect some specific criterions and allow obtaining reproducible data for a large domain of operational parameters. Moreover, they offer a rather rapid way for optimization of the bioreactor and/or upstream or downstream processes, being useful in fermentation scaling-up, too.

The aerobic bioreactor performance can be described and analyzed by means of the mixing time, which allows to establish the optimum hydrodynamic regime into the bioreactor, and by oxygen mass transfer coefficient,  $k_1a$ , which indicates the oxygen transfer rate that is reached for certain fermentation conditions. The mixing time represents one of the most useful criterion for characterization of the mixing intensity. The mass transfer coefficient, k<sub>1</sub>a represents the most important parameter implied on the design and operation of mixing-sparging equipment of the aerobic bioreactors. Both parameters values are affected by a lot of factors, such as geometrical and operational characteristics of the vessels, media composition, type, concentration and microorganisms' morphology, biocatalysts properties (particle diameter, porosity, etc.).<sup>1-9</sup>

For these reasons, the aim of our experiments is to analyze the influences of some specific geometrical and operational parameters of aerobic stirred bioreactors (geometrical characteristics, apparent viscosity or biomass concentration, rotation speed, aeration rate, etc.) on mixing time and oxygen mass transfer coefficient for *Penicillium chrysogenum*, free mycelia and mycelial aggregates or pellets, broths, using a large domain of operating variables. By means of the experimental data some mathematical correlations have been established for the considered systems. The proposed equations allow, unitary and comparatively, analyze the mixing and oxygen transfer efficiency for stirred bioreactors at different fermentation conditions.

## Materials and method

The experiments were carried out in 5 1 (4 1 working volume, ellipsoidal bottom) laboratory bioreactor (Biostat A, B. Braun Biotech International), with computer-controlled and recorded parameters. The bioreactor mixing system consists of two turbine stirrers and three baffles. The bioreactor and impeller characteristics are given in Table 1.

For the experiments on mixing efficiency, the upper stirrer was placed on the shaft at a distance varying between 32 and 128 mm (0.5 d and 2 d) from the lower one. For oxygen mass transfer studies, the distance between the stirrers was maintained at 64 mm (L = d). The rotation speed was varied between 0 and 700 rpm. The experiments were carried out for a Reynolds number intervals

<sup>&</sup>lt;sup>+</sup>for correspondence

d, mm	d/D	H/D	w/d	l/d	h/d	No. N <sub>b</sub> blades	No. N <sub>b</sub> baffles	s/d	$d_{\rm e}/d$	l <sub>e</sub> /d
64	0.36	2.25	0.12	0.28	1	6	3	0.20	0.21	2.81

Table 1 – Characteristics of bioreactor and impeller

below 25,000, which correspond to the laminar, transitory and low turbulent flow regime, and avoid the "cave" formation at the broths surface (for rotation speed over 700 rpm and L = 2 d).

The sparging system consists of a single ring sparger with 64 mm diameter, placed at 15 mm from the vessel bottom, having 14 holes with 1 mm diameter. The air volumetric flow rate was varied from 0 to 450 1 h<sup>-1</sup> (air superficial velocity up to  $5.02 \ 10^{-3} \ {\rm m \ s^{-1}}$ ).

The biomass used consists of fungus (*Penicillium chrysogenum*), with two morphological conformations: free mycelia and mycelial aggregates (pellets, with the average diameter of 1.6 - 1.8 mm); in both cases, the biomass concentration in broths varied between 4 and 33.5 g l<sup>-1</sup> d.w.

Owing to the difficulty of *in-situ* measurement of viscosity during the experiments, the viscosity was measured before and after each experiment using a viscometer of Ostwald type.<sup>10</sup> Both the experiments and viscosity measurements were carried out at a temperature of 21 °C. Any viscosity or morphological conformation change was recorded during the experiments.

The values of mixing time have been determined by means of the tracer method using a solution of 2 mol 1<sup>-1</sup> KOH as tracer, the time needed to the medium pH-value to reach the value corresponding to the considered mixing intensity being recorded.<sup>1</sup> The tracer volume was of 0.5 ml, the tracer being injected opposite to the pH electrode, at 10 mm from the liquid surface. The pH electrode was placed at 20 mm from the vessel bottom. Because the tracer solution density is close to the liquid phase density, the tracer solution flow follows the liquid flow streams and there are no errors due to tracer buoyancy. The pH variations were recorded by bioreactor computer-recorded system. Each experiment has been carried out three or four times, for identical conditions, the average value of mixing time being used. The maximum experimental error was of  $\pm$  3.4 %.

For  $k_1a$  values determination the static method has been used.<sup>2,7,11</sup> This method has the advantages that it can be applied for different media (for establishing the effect of media components on oxygen mass transfer) and does not involve chemical reactions that could affect the measurement precision. The solved oxygen concentrations in broth were measured using an oxygen electrode of InPro 6000 Series type (Mettler Toledo). As it was underlined in literature, because the  $k_1$ a values were in all cases less than 0.3 s<sup>-1</sup>, it was assumed that the response of the oxygen electrode to the change in the oxygen concentration is sufficiently fast and does not affect the determination accuracy.<sup>10–12</sup>

Each experiment has been carried out three or four times, for identical conditions, the average value of oxygen mass transfer coefficient being used. The maximum experimental error was of  $\pm 4.08$  %.

The mathematical correlations, which describe the influences of considered factors on mixing time and  $k_la$  for each microbial culture, were developed on a PC using MATLAB software. For the experimental data, a multiregression analysis was performed. It was chosen a nonlinear equation form that may be liniarized by applying logarithmic function, the difference between the experimental and modeled value being reduced to a minimum. By means of a MATLAB program, the regression coefficients and standard deviation were calculated.

# **Results and discussion**

#### **Mixing time**

Generally, the analysis of mixing efficiency for the aerated mechanical stirred systems is derived from that of non-aerated systems, due to the less complicated flow phenomena for the second ones. But, the prediction of mixing time for aerated broths by means of the equations established for non-aerated systems offers values lower for about 1.2 - 2 times compared with the experimental data.<sup>3</sup> Thus, the aeration influence on mixing efficiency has to be distinctly analyzed.

For numerous fermentations, owing to the decrease in pumping capacity of the stirrer, due to the cavity formation, and to the compartmentalization in regions around each of the stirrers, the aeration increases the mixing time compared with non-aerated systems. However, as it was affirmed in literature, the deviations from the obtained values for non-aerated broths depend on the constructive and functional characteristics of the bioreactor. Thus, the influence of number and position of the stirrers on the shaft is unknown, and the influence of the gas flow rate is different for different rotation speed or Reynolds number values.<sup>6</sup> Furthermore, the most of these models can predict the mixing time values for Re > 10000, this flow regime being rarely reached in the large-scale bioreactors due to the microorganisms' sensitivity to shear stress. For Re < 10000, these models need some corrections.<sup>4</sup>

The differences between the real flow of the aerated broths that are mechanically mixed and the theoretical flow models for aerobic conditions are amplified by the presence and concentration of the biomass. For this reason, the aim of these studies is to analyze and quantify in a unitary manner the influences of *P. chrysogenum* biomass concentration and morphology on the mixing time, in correlation with the geometrical and operational characteristics of the aerobic stirred bioreactor.

The performance of a bioreactor containing a fungus fermentation broth is greatly affected by the rheological properties of the broth. These properties are controlled mainly by the biomass concentration, its growth rate and morphology. Some of the morphological characteristics, such as the geometry (length, diameter, branching frequency) and flexibility of hyphae and the hyphal-hyphal interactions, can influence the mixing efficiency. Generally, the fungus suspensions exhibit a pseudoplastic or Bingham plastic behavior.<sup>13,14</sup> Unlike other microorganisms, the fungus could grow on two morphological conformations: free mycelia and mycelial aggregates (pellets). For both morphological struc-

tures, the accumulation of fungus biomass induces a significant increase of broths viscosity and controls the rheological behavior. The magnitude of this influence depends on the fungus morphology. Thus, for *P. chrysogenum* strains used in these experiments, the apparent viscosity of suspension was of 172.5 mN s m<sup>-2</sup> for free mycelia and 88.4 mN s m<sup>-2</sup> for pellets, at a biomass concentration of 33.5 g l<sup>-1</sup> d.w.

Contrary to the non-aerated media, for which the mixing time is reduced by increasing the rotation speed value, for aerated broths the influence of impeller rotational speed is different and must be related to the apparent viscosity or biomass concentration, air flow rate and distance between the stirrers on the shaft.

As it was stated in the previous papers for aerated water and simulated broths without solid phase, the mixing time initially decreases with rotation speed increasing, reaches a minimum value, increasing then.<sup>3</sup> The value of the rotation speed that corresponds to the minimum of mixing time (*critical rotation speed*) depends on the apparent viscosity and aeration rate. This evolution is the result of the modification of mixing mechanism with the increase of rotation speed in presence of bubbles.

For *P. chrysogenum* broths, besides the common parameters influencing the mixing (rotation speed, air flow rate, impeller geometry, biomass concentration), the fungus morphology exhibits a significant effect. The Figure 1 indicates the exis-



Fig. 1 – Influence of impeller rotation speed on mixing time for P. chrysogenum pellets and free mycelia broths ( $C_X = 33.5 \text{ g } l^{-1} \text{ d.w.}$ ).

tence of a minimum mixing time value for the rotation speed of 500 rpm, this minimum becoming more evident for pellets. The phenomena is due, both, to the lower apparent viscosity of *P. chrysogenum* pellets suspensions compared to free mycelia ones, and to a more pronounced tendency of deposition for pellets.

For the increase of biomass concentration from 4 to 33.5 g  $l^{-1}$  d.w., the rotation speed of 500 rpm and the distance between stirrers of d, the values of mixing time were increased as follows:

- P. chrysogenum pellets:
  - for 18.7 times at 75 1 h<sup>-1</sup> aeration rate
  - for 13.5 times at 400 1 h<sup>-1</sup>
- P. chrysogenum free mycelia:
  - for 15.9 times at 75 1  $h^{-1}$ 
    - for 7.7 times at 400 1  $h^{\!-\!1}$

These results indicate the stronger unfavorable effect of biomass deposition on circulation velocity of air – broth dispersion compared with the effect of apparent viscosity. However, due to the hyphal – hyphal interactions and, consequently, to the higher apparent viscosities, the mixing efficiency for free mycelia broths is about 2 - 3 times lower than that for pellets broths.

Although, the aeration reduces the mixing time compared with the non-aerated system, due to the supplementary contribution of the pneumatic mixing to the broth circulation, the magnitude of this influence strongly depends both on the apparent viscosity of liquid phase, and on the concentration and characteristics of biomass.

Indifferent of the concentration and morphology of *P. chrysogenum*, for stirrer rotation speed below 400 rpm the mixing time decreases with aeration increase.

Thus, for lower *P. chrysogenum* concentrations and both morphological conformations, the mixing time initially increases with aeration rate, reaches a maximum value and decreases. This variation can be explained by formation of small bubbles, due to the presence of solid phase which avoids the bubbles coalescence, those rise being strongly hindered by the high apparent viscosity of fungus broths. After the flooding point the mixing time is reduced owing to the intensification of dispersion circulation (for the impeller speed of 400 rpm the flooding point was reached at an aeration volumetric rate of  $200 - 300 \ 1 \ h^{-1}$  for *P. chrysogenum* pellets, respectively 150 1 h<sup>-1</sup> for free mycelia).

By increasing the biomass concentration the variation of mixing time with aeration rate is gradually changed. Therefore, the continuous reducing of mixing time with the aeration intensification has been obtained for 33.5 g  $l^{-1}$  d.w. *P. chrysogenum*. In these systems, the high apparent viscosity of fungus suspensions controls the mixing efficiency, the mechanical agitation is poor and the relative contribution of pneumatic mixing to the broths circulation is



Fig. 2 – Influence of volumetric air flow rate on mixing time for P. chrysogenum pellets and free mycelia broths at 600 rpm.

important. Owing to the superior apparent viscosity which reduces considerably the relative contribution of mechanical agitation to the broths mixing, these phenomena are more pronounced for *P. chrysogenum* free mycelia. For the studied systems and experimental conditions, the lowest values of mixing time have been reached for a distance of *d* between the stirrers placed on the shaft. The order of the increase of mixing time function of the stirrers' position was as follows:

$$d < 0.5 \ d < 1.5 \ d < 2 \ d$$

The difference between the mixing intensity, induced by placing the stirrers at a distance of d and that obtained for the other positions of the stirrers on the shaft, is indicated in Figure 2. Owing to the solid phase deposition, the distance of 0.5 d or d between stirrers can facilitate the mixing. But, if the stirrers are too close (0.5 d), the bubbles are accumulated and coalescence around the stirrer. The phenomena of air accumulation in the region around the stirrer is amplified at higher biomass concentrations and it reduces the velocity of dispersion circulation.

The cumulated influence of rotation speed and volumetric air flow rate on mixing time at L = d for both morphological conformations of *P. chryso-genum* is plotted in Figure 3.

The experimental data have been included in some mathematical correlations which describe uniquely the influence of aeration, rotation speed and distance between the stirrers on the mixing time for aerobic stirred bioreactors containing fungus broths. The following correlations have been established:

a) fungus (P. chrysogenum) pellets:

$$t_{\rm m} = 0.14 \cdot \frac{\gamma_{\rm x}^{1.21}}{Q_{\rm a}^{0.26} \cdot n^{0.5} \cdot L^{0.50}} \tag{1}$$

b) fungus (P. chrysogenum) free mycelia:

$$t_{\rm m} = 0.36 \cdot \frac{\gamma_{\rm x}^{1.06}}{Q_{\rm a}^{0.36} \cdot n^{0.67} \cdot L^{0.69}}$$
(2)

The proposed models offer a good agreement with the experimental data, the average deviation being of  $\pm 7.2$  % for *P. chrysogenum* pellets and  $\pm 8.1$  % for *P. chrysogenum* free mycelia.

Analyzing the determination coefficients, which represent the square of correlation coefficients for the proposed equations, it can conclude that the considered factors influence the mixing time in an extent of 96.8 % for *P. chrysogenum* pellets and 94.9 % for *P. chrysogenum* free mycelia. The rest of 3.2 %, respectively, 5.1 % can be attributed to the effect of other factors, namely: number, position and geometry of baffles, temperature, etc.



Penicillium chrysogenum free mycelia



Fig. 3 – Cumulated influence of rotation speed and volumetric air flow rate on mixing time for P. chrysogenum pellets and free mycelia broths (L = d).

## Oxygen mass transfer coefficient

## Submerged aeration

The increase of viscosity, as the result of biomass accumulation, induces two direct major effects on oxygen mass transfer: the reduction of turbulence and the perturbation of bubbles dispersion - coalescence equilibrium. This equilibrium is supplementary affected by solid phase, which could amplify or diminish the coalescence process, depending on the concentration and morphological characteristics of microorganisms.

As it can be observed from Figure 4, the increase of P. chrysogenum free mycelia concentration leads to the decrease of k<sub>i</sub>a values. Thus, for the considered variation domain of the main parameters taken into account, respectively, superficial air velocity from 8.36  $10^{-4}$  to 5.02  $10^{-3}$  m/s and specific power input from 100 to 500 W m<sup>-3</sup>, k<sub>1</sub>a was reduced for 3.7 times for  $\gamma_{\rm X}$  increase from 4 to 36.5 g 1<sup>-1</sup> d.w.

These results are in relation with the increase of apparent viscosity of broths corresponding to the variation of biomass concentration between the mentioned limits. Thus, the apparent viscosity of the suspension of fungus mycelial aggregates increased for 44.2 times and fungus free mycelia for 63.9 times for  $\gamma_X$  increase from 4 to 36.5 g l<sup>-1</sup> d.w.<sup>14</sup> Consequently, for the fungus free mycelia broths the influence of power input was diminished by the high viscosity, the values of oxygen mass transfer coefficient being very close for entire domain of aeration rate (Figure 4).

The cultures of P. crysogenum mycelial aggregates exhibit a particular behavior. For these broths, indifferent of bioreactor operating conditions, the increase of biomass concentration initially induces the increase of oxygen mass transfer rate. As it was given in literature, this phenomena was also observed for different types of bioreactors and is explained by the interaction of pellets with bubbles. Thus, bellow a certain biomass concentration level, the pellets increase the turbulence in the film surrounding the air bubbles, promoting the bubbles surface renewal and bubbles disruption.<sup>11,15,16</sup> This effect is attributed especially to small diameter pellets (below 2.5 mm), for larger particles diameters the increase of biomass amount leading to the continuously decrease of oxygen mass transfer coefficient.17-20

The influence of energy dissipated by mechanical mixing is considerable different to that observed for simulated broths without biomass<sup>21</sup>. Thus, contrary to the simulated broths for which the increase of volumic power input leads to the  $k_1a$  increase, for the most of biomass suspensions studied, the mix-





Fig. 4 – Influence of biomass concentration on oxygen mass transfer coefficient ( $v_{\rm S} = 5.02 \ 10^{-3} \ {\rm m \ s^{-1}}$ ).

ing intensification leads to the diminution of oxygen mass transfer rate (Figure 5).

This effect is more pronounced at lower aeration rate and biomass concentration, and is the result of finest dispersion of air, the cells adsorption blocking more easily the bubbles surface. At higher aeration rate, the air dispersion is amplified by combined action of mechanical and pneumatic mixing.

At lower superficial air velocity and higher biomass concentration, the variation of k<sub>1</sub>a with the increase of power consumption reaches a maximum corresponding to 300 W m<sup>-3</sup>, decreasing then. For these systems, the intensification of mixing initially compensates the negative effect of bubbles surface blocking, owing to the redistribution of adsorbed cells and the renewal of gas-liquid interface.

For volumic power input over 300 W m<sup>-3</sup>, the bubble coalescence is diminished, small bubbles are formed, thus increasing the relative importance of blocking effect.

This evolution of oxygen mass transfer coefficient can be correlated with the influence of mechanical agitation intensity on mixing time for the considered fermentation systems. Therefore, as it was underlined in the former studies on mixing time for fungus broths, the power input value of



Fig. 5 – Influence of specific power input on oxygen mass transfer coefficient  $(C_{\chi} = 33.5 \text{ g } l^{-1} \text{ d.w.})$ 

 $300 \text{ W/m}^3$  corresponds to the minimum level of mixing time, consequently to the maximum efficiency of mixing.

Owing to their significant higher apparent viscosities, for fungus free mycelia suspensions the influence of power consumption on  $k_i a$  is attenuated, the relative importance of the influence of mixing intensity becoming lower compared with that of aeration rate or biomass concentration.

According to those above presented, below a certain level of *P. chrysogenum* pellets concentration, the presence of biomass promotes the turbulence and air dispersion into the broths. For this reason, contrary to the earlier results, the increase of biomass amount to 16 g  $l^{-1}$  d.w. amplifies the mixing and, consequently, the oxygen mass transfer (Figure 5).

Although, the intensification of mixing leads to the intensification of oxygen mass transfer, the increase of  $k_1a$  does not counteract the increase of power consumption demand for it. For better characterization of bioreactors performances from the view point of oxygen mass transfer, the term of *oxygen transfer efficiency*,  $E_{O_2}$ , was introduced and defined as:<sup>22</sup>

$$E_{O_2} = \frac{k_1 a}{\frac{P_a}{O}}$$
(3)

Because of the effect of mixing intensity on oxygen mass transfer for biomass suspensions, the reducing of oxygen mass transfer efficiency becomes more significant compared with systems without solid phase. Figure 6 indicates two variation domains of  $E_{O_2}$  with the energy dissipated by mechanical agitation.

Thus, for volumic power input below 200 W m<sup>-3</sup> for fungus cultures with both morphological structures, the oxygen mass transfer efficiency is reduced for about 4 - 7 times by increasing power consumption, the effect that is enhanced by intensifying mixing.

At volumic power consumption over 200 W m<sup>-3</sup>,  $E_{O_2}$  decreases very slowly. Therefore, it can be concluded that, compared with simulated broths, for biomass suspensions the maximum value of oxygen mass transfer rate can be reached at low power consumption.

Aeration rate influences oxygen mass transfer through air hold-up, interfacial area value and media circulation in bioreactor. For low viscosity broths, Newtonian broths or for media containing electrolytes, tensides, polymeric compounds, the bubbles coalescence is avoided, the average bubbles size being reduced. For these systems, the interfacial area between gas and liquid phase is high. But, this positive effect can be diminished due to the presence of biomass, which can reduce the oxyPenicillium chrysogenum mycelial aggregates



Fig. 6 – Influence of specific power input on oxygen mass transfer efficiency ( $C_X = 33.5 \text{ g} \text{ }^{-1} \text{ d.w.}$ )

gen solubility or can obstruct mass transfer by adsorption to the bubbles surface.

For high viscous or non-Newtonian broths, the bubbles exhibit the tendency to coalescence around the stirrers, thus reducing the interfacial aria and leading to a heterogeneous distribution of air into the bioreactor. Furthermore, the cells adsorption to bubbles surface reduces the interfacial area and, therefore, the oxygen mass transfer rate from air to broth. For this reason, a supplementary increase of aeration rate induces the extend of turbulence degree, the homogeneous distribution of air into the broth, and the increase of oxygen concentration gradient between gaseous phase and media. Thus, the experimental results plotted in Figure 7 underlined the positive effect of superficial air velocity on  $k_1a$ .

These data suggest that the favorable influence of turbulence increase by pneumatic agitation counteracts the blocking effect and coalescence increase due to the broth high viscosity and non-Newtonian behavior. Although the aeration influence was the same for all studied systems, the amplitude of this effect is enhanced by viscosity. Modifying the power input between 100 and 500 W m<sup>-3</sup> and superficial air velocity from 8.36 10<sup>-4</sup> to 5.02 10<sup>-3</sup> m s<sup>-1</sup>, the maximum increase of  $k_1a$  of 6.2 times was





Penicillium chrysogenum free mycelia



Fig. 7 – Influence of superficial air velocity on oxygen mass transfer coefficient ( $P_a/V = 500 W m^{-3}$ ).

reached for *P. chrysogenum* free mycelia. The lower relative increase of oxygen mass transfer rate was obtained for *P. chrysogenum* mycelial aggregates (1.7 times) and is the result of higher turbulence induced by solid particles in the liquid film surrounding the bubbles, thus diminishing the effect of pneumatic mixing on turbulence extent.

The cumulated influence of specific power input and superficial air velocity on  $k_1a$  for submerged aerated *P. chrysogenum* pellets and free mycelia broths is plotted in Figure 8.

The experimental data obtained for submerged aeration have been included in some mathematical correlations which describe unitary the influence of biomass concentration, specific power input and superficial air velocity on  $k_1$  for stirred bioreactor containing fungus broths. The general expression of the proposed equations is:

$$k_1 a = \alpha \cdot \gamma_{\rm X}^{\beta} \cdot \left(\frac{P_{\rm a}}{Q}\right)^{\gamma} \cdot v_{\rm S}^{\delta} \tag{4}$$

The influence and the relative importance of the considered variables are suggested by the coefficients a, b, g and d values. The values of these coefficients are specific for each microorganism type



Fig. 8 – Cumulated influence of specific power input and superficial air velocity on oxygen mass transfer coefficient for submerged aerated P. chrysogenum pellets and

or morphology and were calculated by the multiregression method using MATLAB software. Thus, the following correlations have been established:

a) fungus (*P. chrysogenum*) pellets:

$$k_1 a = 0.193 \cdot \frac{v_{\rm S}^{0.257} \cdot \left(\frac{P_{\rm a}}{Q}\right)^{0.0288}}{\gamma_{\rm X}^{0.269}} \tag{5}$$

b) fungus (P. chrysogenum) free mycelia:

$$k_1 a = 33.59 \cdot \frac{v_{\rm S}^{0.94}}{\gamma_{\rm X}^{1.012} \cdot \left(\frac{P_{\rm a}}{Q}\right)^{0.0463}} \tag{6}$$

The proposed models offer a good agreement with the experimental data, the maximum deviation being  $\pm$  8.4 % for *P. chrysogenum* pellets and  $\pm$  7.6 % for *P. chrysogenum* free mycelia.

#### Surface aeration

Oxygen diffusion from gas to liquid phase can simultaneously occur through free surface of broth. The relative magnitude of surface aeration, compared with submerged aeration, becomes more important for small bioreactors or for vessels with H/D ratio close to the unit.<sup>8,22</sup>

The absorption of oxygen at the surface is controlled by liquid turbulence, as well as by media components, which can enhance or reduce the oxygen solubility. Furthermore, for broths containing biomass, the cells adsorption to liquid free surface becomes an important limiting factor for surface aeration.

As it can be observed from Figure 9, the  $k_{la}$  order of magnitude for surface aeration is for two units lower than that obtained for submerged aeration, thus suggesting the negligible contribution of surface aeration to the total oxygen mass transfer for the considered systems.

For the lowest used superficial air velocity (8.36  $10^{-4}$  m/s), the contribution of surface aeration to total oxygen mass transfer was:

- *P. chrysogenum* mycelial aggregates: 0.01 - 0.25 %

-P. chrysogenum free mycelia: 0.01 - 0.25 %.

Compared with simulated broths having similar apparent viscosities, these values indicate the major role of biomass on surface aeration, both by reducing the oxygen solubility in media, and by blocking the liquid surface (the contribution of surface aeration to the total mass transfer of oxygen was for about 15 - 20 times lower than for simulated broths).

For surface aeration, the significant influence of biomass concentration on oxygen mass transfer is in-





Fig. 9 – Influence of specific power input on oxygen mass transfer coefficient for surface aeration.

dicated by the exponential function included in the proposed correlations for  $k_1a$  calculation. The  $k_1a$  variation suggested the following general expression of the equations proposed for surface aeration:

$$k_1 a = \frac{1}{e^{\alpha}} \cdot \left(\frac{P}{Q}\right)^{\beta} \tag{7}$$

where exponents a and b are function of biomass concentration. Using the same multiregression method, the following correlations have been established:

a) fungus (P. chrysogenum) pellets:

$$k_1 a = \frac{1}{e^{12.77 + 0.227 \cdot \ln \gamma_X}} \cdot \left(\frac{P}{Q}\right)^{0.62 - 0.109 \cdot \ln \gamma_X}$$
(8)

b) fungus (P. chrysogenum) free mycelia:

$$k_1 a = \frac{1}{e^{11.46 + 0.729 \ln \gamma_X}} \cdot \left(\frac{P}{Q}\right)^{0.093 + 0.057 \cdot \ln \gamma_X} \tag{9}$$

The maximum deviations from the experimental data of the k<sub>1</sub>a values calculated with the equations (8) and (9) are  $\pm 9.1$  % for *P. chrysogenum* pellets and  $\pm 8.6$  % for *P. chrysogenum* free mycelia.

## Conclusions

The mixing time and oxygen mass transfer coefficient are the most useful criterions for evaluate and model the performances of the aerobic stirred bioreactors, for its optimization and scaling-up. By studying the mixing time and oxygen mass transfer for fungus broths (*Penicillium chrysogenum* mycelial aggregates and free mycelia) in an aerobic stirred bioreactor, the following conclusions can be drawn:

1. The variation of mixing time with impeller rotation speed indicated the existence of the critical rotation speed (500 rpm) that corresponds to the maximum of mixing intensity. The most efficient mixing was obtained for a distance between the stirrers on the shaft equal with their diameters.

2. Compared with simulated broths without biomass, but having the same apparent viscosities, the oxygen mass transfer rate for biomass suspensions was lower. This evolution of  $k_1a$  is the result of bubbles surface blocking by cells adsorption. The magnitude of blocking effect depends on microorganism type, becoming significant for *P. chrysogenum* free mycelia.

The mixing intensification leads to the reducing of  $k_1a$ . This effect is more pronounced at lower aeration rate and biomass concentrations, being the results of finest dispersion of air, and, consequently, of easy adsorption of the cells on the bubbles surface. The value of specific power input of 300 W/m<sup>3</sup> corresponds to the minimum level of mixing time, and, therefore, to the maximum rate of oxygen transfer.

3. Compared with simulated broths having similar apparent viscosities, the experiments indicate the major role of fungus biomass on surface aeration, both, by reducing the oxygen solubility in media, and by blocking the liquid surface (for the lowest used superficial air velocity  $8.36 \ 10^{-4} \ m \ s^{-1}$ , the contribution of surface aeration to the total mass transfer of oxygen was for about 15 - 20 times lower than for simulated broths).

#### Notations

 $\gamma_{\rm X}$  – biomass concentration, g l<sup>-1</sup> dry weight

- d stirrer diameter, mm
- $d_{\rm e}$  pH electrode diameter, mm
- D bioreactor diameter, mm
- $E_{\Omega_2}$  oxygen mass transfer efficiency, m<sup>3</sup> J<sup>-1</sup>
- *h* distance from the inferior stirrer to the bioreactor bottom, mm
- H bioreactor height, mm
- l impeller blade length, mm
- $l_{\rm e}$  pH electrode immersed length, mm
- L distance between the stirrers, mm

- n impeller rotation speed, rpm
- *P* power consumption for mixing of non-aerated broths, W
- $P_{\rm a}$  power consumption for mixing of aerated broths, W
- $(P_a/Q)$  specific power input, W m<sup>-3</sup>
- s baffle width, mm
- $t_{\rm m}$  mixing time, s
- $v_{\rm S}$  superficial air velocity, m s<sup>-1</sup>
- $Q_{\rm a}$  volumetric air flow rate, m<sup>3</sup> s<sup>-1</sup>
- w impeller blade height, mm.

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