Measurement of Mass Transfer Coefficients in Airlift Reactors with Internal Loop Using the Glucose Oxidase Method

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This study deals with the measurement of volumetric mass transfer coefficients (k_La) in the internal loop airlift reactors (ILALRs) of different volumes (12 dm^3, 40 dm^3 and 200 dm^3) using the glucose oxidase method. The experimental results of this work were compared with those k_La obtained for the same ILALRs by applying different other methods. Results of this work showed a good agreement with the published results obtained by applying other methods on the non-coalescent medium. The initial glucose mass concentration in the non-coalescent medium was 150 g dm^{-3}. Based on this work, the values of the empirical parameter in the correlation of k_La vs. the total gas hold-up, proposed by *Bello* et al.,[1] were established.

Key words:
Volumetric mass transfer coefficient, internal loop airlift reactor, glucose oxidase method

Introduction

One of the most important substrates for aerobic bioprocesses is oxygen. The cheapest source of oxygen is air. However, microorganisms are able to utilise only dissolved oxygen in a liquid medium. This fact calls for knowledge of oxygen transfer from the gas phase to the liquid phase, which is necessary in designing new bioreactors or providing better process conditions for existing reactors. The volumetric mass transfer coefficient (k_La) is defined as the rate of gas transfer across the gas-liquid interface, per unit volume of the liquid and per unit driving force.

The value of k_La can be measured by applying different methods. Generally, these methods can be divided into two basic groups: direct and indirect methods. The principle of these methods lies in their (bio)chemical or physical features, which determines their application.[2]

In this work the k_La values were measured using the glucose oxidase method (GODM). This method belongs to the group of indirect, biochemical and steady state methods.[2–4] The advantage of the GODM over the other methods (e. g. sulphite oxidation method or hydrazine method) is that properties of the liquid are the same or very similar to the properties of a liquid medium used in fermentation or enzymatic reaction processes. When this enzymatic oxidation method is applied, the k_La values are determined from the rate of gluconic acid production. The mechanism of biocatalytic glucose transformation to gluconic acid involves two steps:

$$\text{glucose} + O_2 \xrightarrow[H_2O]{\text{GOD}} \text{gluconic acid} + H_2O_2 \quad (1)$$

$$H_2O_2 \xrightarrow{\text{CAT}} H_2O + 1/2\ O_2 \quad (2)$$

where GOD is glucose oxidase and CAT is catalase. The overall reaction can be written as follows:

$$\text{glucose} + 1/2\ O_2 \xrightarrow[H_2O]{\text{GOD CAT}} \text{gluconic acid} + H_2O \quad (3)$$

As found by *Nakao* et al.[3] for application of the steady state method, the enzymatic reaction of glucose oxidation is so specific to the substrate (e. g. oxygen and glucose) that its rate is highly reproducible even in the presence of various foreign substrates. Moreover, it was established that, the reaction capacity of the system must exceed the oxygen absorption rate to perform reliable measurements of k_La. This can be achieved when the concentration of glucose is sufficiently above the limitation concentration of glucose for the enzymatic reaction.

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Determination of the volumetric mass transfer coefficient (k_La)

The determination of the k_La values in airlift reactors (ALRs) was based on the measured quasi-steady state concentration of dissolved oxygen and the estimated rate of gluconic acid production. The oxygen balance in the system, where the dissolved oxygen is consumed only by enzymatic oxidation of glucose to gluconic acid, can be written as follows:

$$\frac{dc_0}{dt} = \text{OTR} - \text{OUR} = k_L a(c_0^* - c_0) - q_0 c_E \quad (4)$$

In the quasi-steady state the left hand side of eq. (4) is equal to zero. Then, from the stoichiometry of glucose to gluconic acid conversion (eq. (3)) and the rate of gluconic acid production, the following equation for the k_La values determination can be delivered:

$$k_L a = \frac{r_{Glu}(M_{O_2}/M_{Glu})}{2(c_0^* - c_0)} \quad (5)$$

The gas phase is assumed to be perfectly mixed.

Correlation of the volumetric mass transfer coefficient (k_La) and the gas hold-up (ε_G)

According to *Bello* et al.,[1] under the assumption that the contribution of the gas hold-up in the downcomer is negligible in comparison to that in the riser, the overall mass transfer coefficient could be described as a function of the gas hold-up, i.e. as follows:

$$k_L a = K\left(1 + \frac{A_D}{A_R}\right)^{-1.4+n+0.6+m} \varepsilon_{GR}^{1.6-n} \quad (6)$$

Bello et al. set the exponent, (1.6-n), on the term of ε_{GR} to be equal to 1.2, which was in agreement with the work of *Calderbank*.[5] Moreover it has been shown, that the variation of the k_La values *de facto* is independent of the ratio A_D/A_R, for both the bubble columns and the ALRs, whereas is dependent on the gas hold-up.

However, it is known that in some cases the gas hold-up in the downcomer cannot be negligible. In such cases, a significant amount of mass transfer from the gas phase to the liquid phase can occur in this part of the ALR. Hence, in the present work the determination of the k_La values correlated with the overall gas hold-up was studied. Therefore, eq. (6) can be then rewritten in the following form:

$$k_L a = K(\varepsilon_{GC})^{\alpha} \quad (7)$$

Experimental and methods

The experimental set-up is schematically presented in Fig 1. The measurements were carried out in three internal loop airlift reactors (ILALRs) of different volumes. The reactors were made of glass, the bottom with the gas sparger being made of stainless steel. The total working volumes of the single columns were 12 dm^3, 40 dm^3 and 200 dm^3. The three reactors were of similar geometry to avoid its influence on the hydrodynamic parameters. The chosen similarity criteria were: column slightness (ratio of column height to column diameter) and the ratio of cross-sectional areas of the downcomer and the riser (A_D /A_R). However, the two smaller ALRs (12 and 40 dm^3) had an enlarged degassing zone and the 200 dm^3 ALR had a simple separator of the same diameter (D_S) as that of the column (D_C) (see Tab. 1).

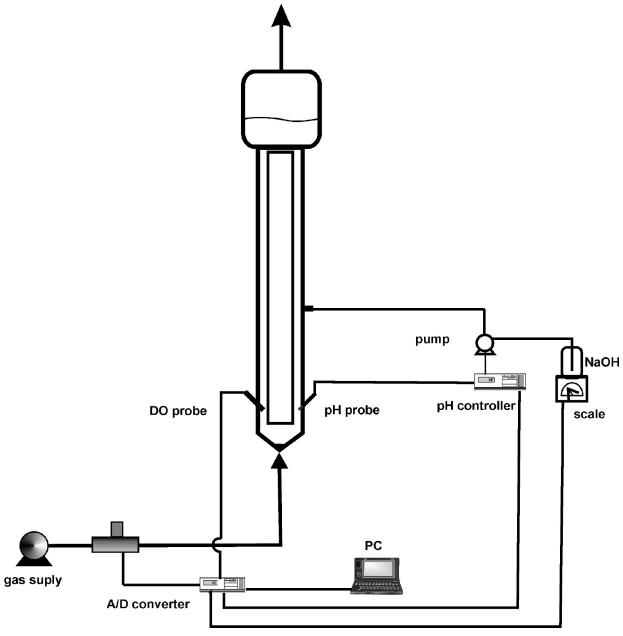


Fig. 1 – *Experimental set-up*

The geometrical details of the used ILALRs are listed in Tab 1. In all measurements, only the internal tube was sparged, whereby the gas sparger was located at the bottom in form of a perforated plate made of teflon or stainless steel. The details of the gas spargers used in this work are given in Tab 2.

In the reactors with working volumes of V = 12 dm^3 and 40 dm^3 the gas input was controlled

Table 1 – *Geometrical details of the reactors used*

Working volume V/dm^3	D_C / m	H_L / m	H_R / m	D_R / m	D_S / m	H_B / m	A_D/A_R / –	H_L/D_C / –
12	0.108	1.34	1.145	0.070	0.157	0.030	1.23	12.4
40	0.157	1.93	1.710	0.106	0.294	0.046	0.95	12.3
200	0.294	3.10	2.700	0.200	0.294	0.061	1.01	10.5

Table 2 – *Types of gas spargers used*

Working volume V/dm^3	Material	Number of holes	Hole diameter d/mm
12	Teflon	25	0.5
40	Teflon	50	0.5
200	stainless steel	90	1.0

by a rotameter. If a higher gas input was required (V = 40 dm^3 and 200 dm^3 reactors), the gas input was controlled by a mass flow controller (BROOKS-5853E). All experiments were carried out at the temperature of 30 ± 0.3 °C under atmospheric pressure. Both the oxygen probe (InPro 6800, Mettler Toledo) and the pH sensor (405-DPAS-SC-K8S, Mettler Toledo) were placed at the bottom of the downcomer. The value of pH was kept at 5.5 ± 0.2 using a solution of NaOH. A c = 12 mol dm^{-3} solution of NaOH was used for experiments carried out in the 40 dm^3 and 200 dm^3 ALRs whereas a c = 6 mol dm^{-3} solution of NaOH was used for the 12 dm^3 ALR. Data acquisition of all process parameters (pH, DO, inlet pressure and temperature of gas, process temperature and the amount of added NaOH) were done by an A/D converter (API, Slovakia) connected to a PC. A more detailed description of the experimental arrangement can be found in our previous papers.[6,7]

Air was used as the gas phase and the liquid phase consisted of deionized water and glucose (initial mass concentration of γ = 150 g dm^{-3}). For one experiment in the 12 dm^3 ALR, pure oxygen was used as the gas phase. The enzyme applied in this work had the following specification: 1 g of enzyme powder consisted of 10 000 glucose oxidase units and had a sufficient level of catalase activity (commercial name: Gluzyme 10 000BG, Novozymes Denmark). The concentration of the dissolved enzyme powder was γ = 1 g dm^{-3}.

To determine the k_La values (eq. (5)), a time profile of the gluconic acid concentration during the experiment should be known. The amount of gluconic acid produced can be directly calculated from the actual amount of NaOH necessary for neutralisation, keeping the pH at a value of 5.5.

Results and discussion

To estimate the equilibrium dissolved oxygen concentration, c_0^*, the composition of the liquid phase should be taken into account.[8,9]

Generally, it is known that an ALR consists of four main parts: riser, downcomer, degassing zone and bottom section. Each part of the ALR can be described with different hydrodynamic behaviour. Liquid or (solid-) gas-liquid dispersion circulates through the parts of ALR. The correct way to measure the k_La values for such a type of circulation reactor should be by determination of k_La values for each part of the reactor separately. However, it is possible to consider the reactor as one unit if the following criterion is fulfilled.[10]

$$k_L a\, t_c \leq 2 \tag{8}$$

where t_c is the circulating time.

In accordance to the values of circulation velocity attained in the reactors described previously[11] and the oxygen transfer rate observed in the present work, criterion was sufficiently fulfilled. In present case, the highest value of the term on the left hand side of eq. (8) was 0.8.

All experiments started with an initial glucose mass concentration of 150 g dm^{-3} without the presence of any gluconic acid and NaOH at a considered gas flow rate. After a certain time of quasi-steady state of the enzymatic reaction, the gas flow rate was changed to different values to measure k_La. Hence, all determinations of k_La values for one ILALR were done in one run. That means that the concentrations of glucose, gluconic acid and Na^+ were changing approximately from 150 g dm^{-3} to 100 g dm^{-3} for glucose and from 0 g dm^{-3} to 55 g dm^{-3} for gluconic acid. Solution of NaOH was titrated to keep the constant pH value. Hence, mass concentration of Na^+ increased from 0 g dm^{-3} to about 11 g dm^{-3} during one run. To verify the measured values and the influence of medium composition on them, at the end of each run, the gas flow rates were switched to the initial values to compare the k_La values obtained with the initial values. Moreover, new experimental runs were carried out by using different initial gas flow rates. Practically, almost the same measured values of k_La were monitored at the beginning and at the end of the process. The same fact was observed using different initial gas flow rates. That indicates that a change of the medium composition in this range has no signifi-

cant effect on the oxygen mass transport from the gas phase to the liquid phase.

The results of k_La measurements confirmed a positive influence of gas flow rate on the intensity of oxygen mass transfer. Almost a linear experimental dependency of the k_La values on the superficial gas velocity (u_{GC}) was reported for the whole gas flow rate range applied in this work (Fig. 2). It can be noted that the composition of gas has no influence on the values of k_La (see experimental results referring to the 12 dm^3 ILALR with air and oxygen as the gas phase in Fig. 2).

The mass transfer from the gas phase to the liquid phase is strongly influenced by the total gas hold-up (ε_{GC}) in the dispersion. Fig. 3 shows the effect of u_{GC} on ε_{GC}. The values of ε_{GC} were calculated from the total volume of the liquid and the deduced volume of dispersion. The error of ε_{GC} determination was up to 5 % due to an unsteady dispersion height level and formation of foam.

Fig. 4. shows the variation of the k_La values with ε_{GC} in the ALRs. The experimental dependencies were described by eq. (7). Parameters of the correlation are given in Tab. 3. When the original values of the parameters proposed by *Bello* were used, correlation (7) described all experimental k_La values from all ILALRs with the correlation coefficient of 0.93. When the K parameter was fitted to all experimental k_La values from all ILALRs, its value was 0.61 s^{-1} and correlation (7) described the experimental data with the correlation coefficient of 0.92.

The comparison of the experimental dependency of the k_La values on u_{GC} established in this work (determined using free enzymes in the 12 dm^3 ILALR) with the dependencies obtained by using real biomass of *Aspergillus niger* which was in a pellet form,[7] some differences can be observed (Fig. 5). It was supposed that the values obtained using biomass should be lower due to intracellular glucose oxidase and catalase enzymes. Hence, to convert glucose into gluconic acid, oxygen has to be transferred from the gas phase to the liquid phase and consequently moved by diffusion inside the biomass pellet and then transferred into particular cell. All this events should lead to lower values of

Table 3 – *Values of parameters in eq. (7)*

Parameters	Values proposed by *Bello* et al.[1]	This work
K, s^{-1}	0.47	0.61
α, –	1.2	1.2
R^2	0.93	0.92

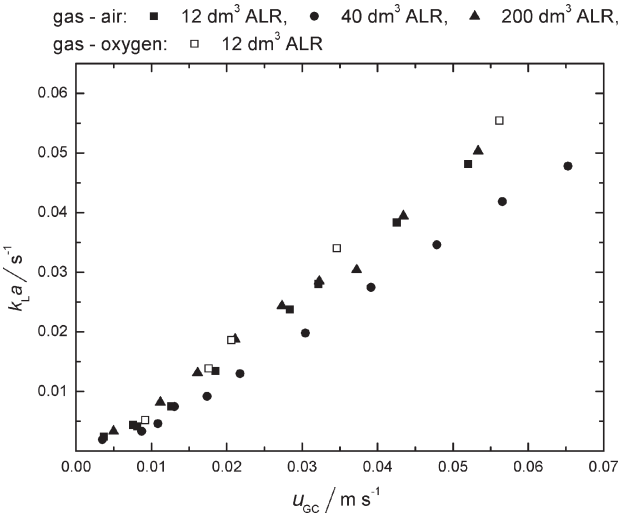


Fig. 2 – *Experimental dependency of k_La values on u_{GC}*

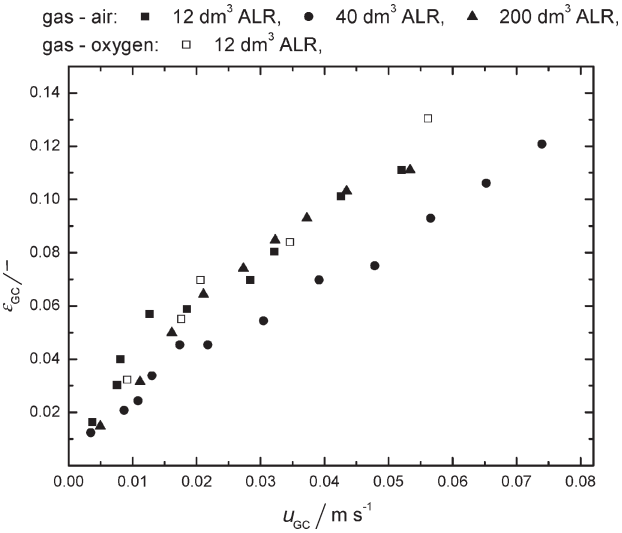


Fig. 3 – *Experimental dependency of ε_{GC} values on u_{GC}*

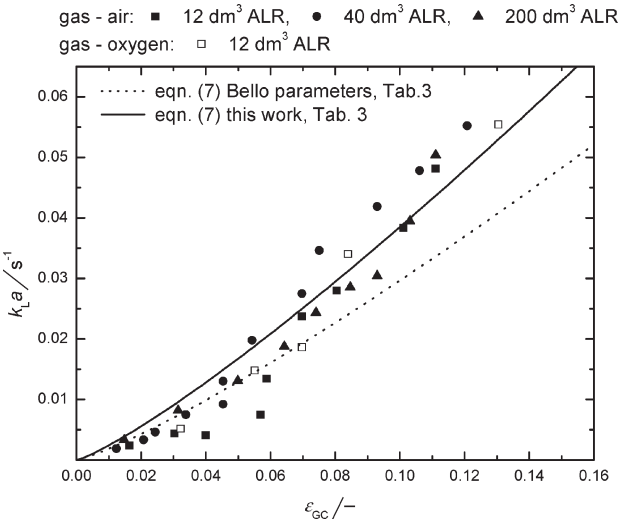
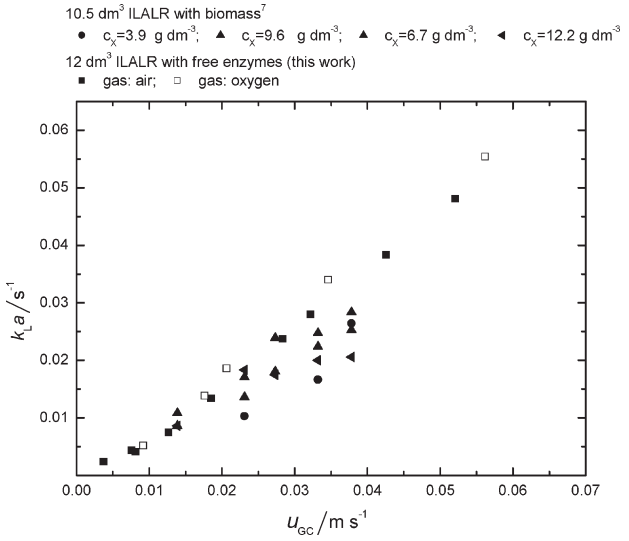
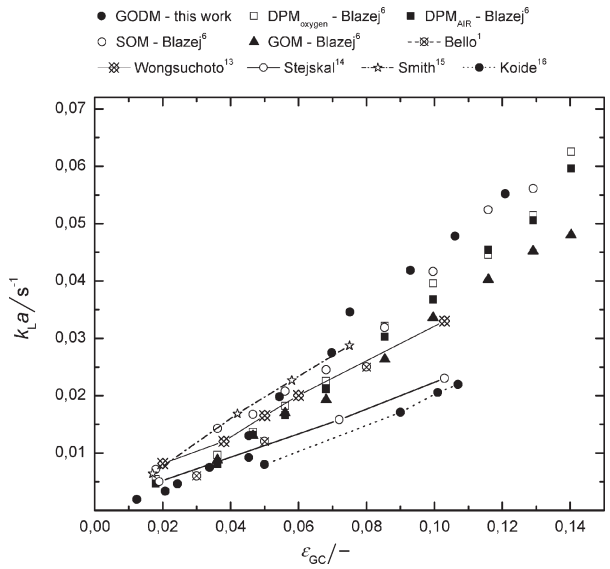


Fig. 4 – *Dependency of k_La values on ε_{GC}*



F i g . 5 – *Comparison of the k_La values obtained in this work with the k_La values measured in the presence of biomass with intracellular enzymes*[7]



F i g . 6 – *Comparison of the k_La values obtained in this work with the k_La values measured applying other methods in the same ILALR*[6] *and with the result of other authors carried out under similar condition*[1,13–16]

the mass transfer coefficient than using free enzymatic reactions.

However, the differences which can be observed in Fig. 5, are not significant. That might be the consequence of the differences in working volumes used in this work what has an influence on hydrodynamics of the system. The experiments of this study and those of the study with biomass[7] were carried out in the same ILAR, but they refer to different working volumes: 12 dm^3 in this study and 10.5 dm^3 in the work applying biomass. Therefore, it follows that the degassing zones of ILALRs differed in volumes of gas-liquid (-solid) dispersion. During the experiments with biomass, a smaller volume of the gas-liquid-solid dispersion was occupying the degassing zone. That led to a lower degassing efficiency of the zone.[12] This fact justifies the higher overall gas hold-ups in the ILALR with working volume of 10.5 dm^3 in comparison with the system using 12 dm^3 of the working volume. Hence, the overall mass transfer in the former system is higher than that of the latter for the given air flow rates.

Fig. 6 refers to the comparison of k_La values measured in this work in the 40 dm^3 ILALR with k_La values determined in a non-coalescent medium by different methods using the same ILALR.[6] The non-coalescent medium in the work of *Blažej* et al.[6] consisted of a 0.3 mol dm^{-3} aqueous solution of Na_2SO_3 for the gassing-out method (GOM) and the dynamic pressure-step method (DPM). Applying the sulfite oxidation method (SOM) a catalyst ($CuSO_4$) was added. The experimental results of this work using the glucose oxidase method (GODM) are in a good agreement with the results obtained by other methods in the non-coalescent medium. Moreover, in Fig. 6 a comparison of our experimental k_La values with the results of other authors[1,13–16] carried out under similar condition is done.

Conclusions

This experimental study deals with the measurements of the volumetric mass transfer coefficients (k_La) in three scales of geometrically similar internal loop airlift reactors by applying the glucose oxidase method. The liquid medium consisted of glucose (mass concentration from γ = 150 g dm^{-3} to approximately γ = 100 g dm^{-3}), gluconic acid (mass concentration from γ = 0 g dm^{-3} to approximately γ = 55 g dm^{-3}) and Na^+ (mass concentration up to γ = 11 g dm^{-3}). The experiments showed that the composition of liquid in these ranges has no influence on the k_La values.

When the semiempirical correlation proposed by *Bello* et al.[1] was used for the prediction of the experimental data applying original values or the fitted value of the K parameter, the correlation coefficients close to one indicated the sufficient description of the experimental k_La values.

The experimental results of this work were compared with the k_La values previously obtained in the same ILALR by different methods. Based on these observations, a further study should be focused on the influence of biomass on k_La determined by the dynamic pressure-step method and glucose oxidase method using inhibited or non-inhibited intracellular glucose oxidase enzymes.

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Notation

A – cross section area, m^2
c – concentration, mol dm^{-3}
D – diameter, m
H – height, m
K – coefficient in eqs. 6 and 7, s^{-1}
k_La – volumetric mass transfer coefficient, s^{-1}
m – exponent in eq. 6, –
M – molar mass, g mol^{-1}
n – exponent in eq. 6, –
q_O – specific oxygen uptake rate
R^2 – correlation coefficients, –
r_{Glu} – gluconic acid production rate, g m^{-3} s^{-1}
t_c – circulating time, s
u – superficial gas velocity, m s^{-1}
V – volume, dm^3

Greek letters

α – exponent in eq. 7, –
γ – mass concentration, g dm^{-3}
ε – gas hold-up, –

Subscripts and superscript

B – distance from the bottom of ALR to the beginning of the riser
C – column
D – downcomer
E – enzyme
G – gas
Glu – gluconic acid
L – liquid
O – oxygen
R – riser
S – separator
X – biomass
* – equilibrium

Abbreviations

ALR – airlift reactor
CAT – catalase enzyme
DPM – dynamic pressure-step method
GOD – glucose oxidase enzyme
GODM – glucose oxidase method
GOM – gassing-out method
ILALR – internal loop airlift reactor
OTR – oxygen transfer rate
OUR – oxygen uptake rate
SOM – sulphite oxidation method

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