

Correlation between Volumetric Oxygen Transfer Coefficient and Power Requirement in Citric Acid Fermentation by *Aspergillus niger*

M.A. HASSAN, N.D. NIK SIN, B. ABDUL GHANI and M.I. ABDUL KARIM

Department of Biotechnology
Faculty of Food Science and Biotechnology
Universiti Pertanian Malaysia
43400 UPM Serdang, Selangor, Malaysia

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ABSTRAK

Satu sistem kultur sesekelompok bagi penghasilan asid sitrik oleh *Aspergillus niger* TISTR 3089 telah dikaji untuk melihat kesan pengudaraan dan pengadukan keatas pekali pemindahan oksigen isipadu ($k_L a$) dan penggunaan kuasa. Tangki berpengaduk 2 liter digunakan dengan halaju pengaduk 500-900 psm dan kadar pengudaraan 0.65 iim dengan glukosa sebagai sumber karbon. $k_L a$ dan penggunaan kuasa berubah dengan tempoh fermentasi. Data yang diperolehi dipadankan dengan korelasi matematik antara $k_L a$ dan keperluan kuasa semasa pengassan bagi sunit isipadu cecair (P_g/V) dan halaju superfisial gas yang dilaporkan oleh penyelidik terdahulu. Dengan memasukkan halaju pengaduk, korelasi-korelasi yang lebih baik didapati, menandakan kepentingan halaju pengaduk di dalam korelasi berkenaan. Perhubungan yang dicadangkan ialah: $k_L a = k (P_g/V)^{0.95} U^{0.67} N^{0.5}$, dimana U ialah halaju superfisial gas dan N ialah halaju putaran pengaduk.

ABSTRACT

A batch culture system for the production of citric acid by *Aspergillus niger* TISTR 3089 was studied to determine the effect of aeration and mixing on the volumetric oxygen transfer coefficient ($k_L a$) and power requirement. A 2-l batch stirred-tank was used with impeller speeds of 500-900 rpm at an aeration rate of 0.65 vvm with glucose as the carbon source. $k_L a$ and power consumption varied with duration of fermentation. The data obtained were fitted to mathematical correlations between $k_L a$ and power requirement during gassing per unit volume of liquid (P_g/V) and superficial gas velocity reported by previous researchers. By including the agitator speed, better correlations were obtained, indicating the importance of stirrer speed in such correlations. The proposed relationship is: $k_L a = k (P_g/V)^{0.95} U^{0.67} N^{0.5}$, where U is the superficial gas velocity and N is the stirrer speed.

INTRODUCTION

Citric acid fermentation is usually done in a batch system, using moulds such as *Aspergillus niger*. During the fermentation process, the effect of environmental factors such as pH, temperature, aeration and mixing are critical (Berry *et al.* 1977). The degree of mixing significantly influences the efficiency of oxygen transfer, as the bubble size can be reduced and gas hold-up increased. Power consumption is closely related to degree of mixing. The relationship between volumetric oxygen transfer

coefficient and power consumption is useful and important in the design and scaling-up of bioreactors. Various correlations have been suggested, especially for newtonian liquids (Cooper *et al.* 1944; Bartholomew 1960; Richards 1961; van't Riet 1983) and non-newtonian systems (Blakebrough and Sambamurthy 1966; Manfredini and Cavallera 1983; Kargi and Moo-Young 1985). Taguchi and Humphrey (1966) studied the relationship between oxygen transfer rate and power consumption in an *Endomyces* fermentation system which is pseudoplastic. The

objective of this study is to investigate the relationship between power consumption and oxygen transfer in citric acid fermentation by *Aspergillus niger*.

MATERIALS AND METHODS

Microorganism

The microorganism used was *Aspergillus niger* TISTR 3089 obtained from Scientific and Technological Research Institute, Thailand. The culture was grown in potato dextrose agar (PDA) slants and kept at 4°C.

Medium

The medium was made up of 180 g/l glucose, 2.0 g/l ammonium nitrate, 2.0 g/l potassium hydrogen phosphate, 0.5 g/l magnesium sulphate, 0.1 mg/l ferric sulphate, 0.1 mg/l zinc sulphate and 0.06 mg/l copper sulphate. The pH was adjusted to pH 4.5. and the medium was autoclaved at 121°C for 15 minutes. Glucose was autoclaved separately and added later.

Inoculum

The culture from the agar slant was transferred to a petri dish containing PDA, and incubated at 30°C for 7-10 days. Sterile water was added to the petri dish containing the spores. The spore suspension was then collected aseptically and its optical density was determined at 565 nm to be within 0.8-0.85.

The spore suspension was then transferred to a shake flask containing the fermentation medium. It was incubated on an orbital shaker at 200 rpm and 30°C for 1-2 days to obtain the inoculum for the fermenter; 10% (v/v) inoculum was used.

Fermentation

The fermenter used was a 2-l Braun Biostat M stirred-tank reactor with a 6-bladed Rushton turbine impeller. The reactor was equipped with ports for air inlet and outlet, acid and alkali, antifoam, inoculation, sampling and pH and oxygen probes.

Pure oxygen was used for aeration. Impeller speeds ranged from 500-900 rpm. Sterile silicone oil was used to control foaming. Fermentation proceeded for 7-8 days. Sampling was done daily. Power requirement, $k_L a$, pH, glucose, citric acid and dry cell weight were determined throughout the fermentation process.

Glucose was determined by the dinitrosalicylic acid method (Miller 1959). Citric acid was determined by HPLC using Lichrosorb RP-18 column with 8 mM sulphuric acid as the mobile phase and UV detector at 210 nm. For the dry weight, the sample was centrifuged, re-suspended with distilled water and centrifuged again before drying in an oven at 105°C overnight. The $k_L a$ was measured by the dynamic gassing-out technique. A torsion dynamometer was used for power measurement.

RESULTS AND DISCUSSION

Table 1 shows the effect of stirrer speed on biomass (cell weight), citric acid production, glucose consumption, pH changes, $k_L a$ and power consumption. The kinetic data showed the expected pattern for any batch fermentation, although the citric acid yield is rather low.

Table 2 compares the experimental fit of correlations based on current experimental data in the literature as well as the new correlation proposed. Using the correlation suggested by Cooper *et al.* (1944), i.e. $k_L a = k (P_g/V)^{0.95} U^{0.67}$, the regression coefficient R^2 is 0.78. When the square root of the stirrer speed is added inside the correlation, a better fit is obtained, with $R^2 = 0.83$.

Richards (1961) suggested $k_L a = k (P_g/V)^{0.4} U^{0.5}$; using 0.4 as the exponent on the power per unit volume. Fitting the current experimental data using that relationship, $R^2 = 0.78$. By including the term for the stirrer speed $N^{0.5}$, there was better correlation with $R^2 = 0.82$.

Taguchi *et al.* (1968) suggested the correlation $k_L a = k (P_g/V)^{0.33} U^{0.56}$. Using this relationship, $R^2 = 0.77$. When $N^{0.5}$ is included inside the relationship, again a better fit of the current experimental data is obtained ($R^2 = 0.81$). This further reinforces the suggestion that the stirrer speed is an important factor in the correlation.

Richards (1961) showed that the liquid mass transfer coefficient (k_L) is proportional to the square root of the stirrer speed. From Calderbank (1967), under constant surface tension and terminal gas velocity, the volumetric gas-liquid interfacial area (a) would be proportional to $(P_g/V)^{0.4} U^{0.5}$. Combining these two for an expression for the volumetric oxygen transfer coefficient, $k_L a$ is then proportional to $(P_g/V)^{0.4} U^{0.5} N^{0.5}$. This gives the theoretical foundation for the proposed correlation in this study.

TABLE 1
 Volumetric oxygen transfer coefficient, power and kinetic data for citric acid fermentation by *Aspergillus niger* TISTR 3089

| Stirrer Speed (rpm) | Time (days) | $k_L a$ (/h) | Power (kW) | Glucose (g/l) | Citric Acid (g/l) | Cell Wt. (g/l) | pH |
|---------------------|-------------|--------------|------------|---------------|-------------------|----------------|-----|
| 500 | 0 | 1.69 | 0.44 | 180 | 0.0 | 1.7 | 3.0 |
| | 1 | 1.73 | 0.70 | 165 | 0.0 | 6.5 | 1.9 |
| | 2 | 1.78 | 0.79 | 140 | 0.6 | 10.4 | 1.8 |
| | 3 | 2.10 | 1.01 | 99 | 2.3 | 12.4 | 1.7 |
| | 4 | 2.21 | 1.14 | 95 | 3.4 | 14.3 | 1.4 |
| | 5 | 2.69 | 1.27 | 88 | 4.2 | 17.1 | 1.4 |
| | 6 | 3.11 | 1.40 | 70 | 4.6 | 17.3 | 1.4 |
| | 7 | 3.38 | 1.44 | 39 | 4.7 | 18.4 | 1.4 |
| 700 | 8 | 4.01 | 1.44 | 21 | 4.8 | 18.6 | 1.4 |
| | 0 | 1.89 | 0.61 | 180 | 0.0 | 1.8 | 3.1 |
| | 1 | 2.26 | 0.80 | 161 | 0.0 | 8.9 | 2.2 |
| | 2 | 3.81 | 1.04 | 130 | 0.8 | 12.3 | 2.0 |
| | 3 | 4.93 | 1.23 | 88 | 2.9 | 14.8 | 1.9 |
| | 4 | 5.24 | 1.41 | 57 | 3.8 | 16.6 | 1.8 |
| | 5 | 5.54 | 1.65 | 56 | 4.7 | 18.5 | 1.8 |
| | 6 | 5.89 | 1.84 | 45 | 5.4 | 20.6 | 1.8 |
| 900 | 7 | 5.99 | 1.86 | 33 | 5.9 | 22.4 | 1.8 |
| | 8 | 6.01 | 1.90 | 20 | 5.9 | 22.4 | 1.8 |
| | 0 | 1.97 | 0.79 | 180 | 0.0 | 1.8 | 3.1 |
| | 1 | 2.56 | 0.79 | 159 | 0.0 | 9.9 | 2.7 |
| | 2 | 3.42 | 0.87 | 140 | 0.9 | 13.9 | 2.5 |
| | 3 | 3.94 | 1.02 | 85 | 3.0 | 16.5 | 2.2 |
| | 4 | 4.75 | 1.26 | 47 | 3.9 | 20.0 | 2.1 |
| | 5 | 5.88 | 1.58 | 25 | 4.9 | 25.4 | 2.0 |
| 6 | 6.11 | 1.97 | 18 | 5.8 | 25.8 | 2.0 | |
| 7 | 6.13 | 2.36 | 16 | 6.1 | 25.8 | 1.9 | |
| 8 | 6.14 | 2.36 | 15 | 6.2 | 25.7 | 1.9 | |

TABLE 2
 Experimental fit of current results to various correlations

| | | R^2 | |
|----|------------------------------|---|------|
| 1. | Cooper <i>et al.</i> (1944) | $k_L a = k (P_g/V)^{0.95} U^{0.67}$ | 0.77 |
| | New correlation | $k_L a = k (P_g/V)^{0.95} U^{0.67} N^{0.5}$ | 0.83 |
| 2. | Richards (1961) | $k_L a = k (P_g/V)^{0.4} U^{0.5}$ | 0.78 |
| | New correlation | $k_L a = k (P_g/V)^{0.4} U^{0.5} N^{0.5}$ | 0.82 |
| 3. | Taguchi <i>et al.</i> (1968) | $k_L a = k (P_g^{0.33}/V) U^{0.56}$ | 0.77 |
| | New correlation | $k_L a = k (P_g^{0.33}/V) U^{0.56} N^{0.5}$ | 0.81 |

CONCLUSION

The results of this study show that in all cases, by including the square root of the stirrer speed inside the equations for correlations suggested by previous researchers, a better correlation between the volumetric oxygen transfer coefficient and power requirement during citric acid fermentation by *Aspergillus niger* was obtained. Thus it is suggested that the stirrer speed should be taken into consideration in such correlations.

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