RESEARCH PAPER
Asian Journal of Bio Science, Vol. 4 Issue 1 : 63-68 (April to September, 2009)

Effect of aeration and agitation on volumetric oxygen transfer in *Pichia pastoris* culture system

S. ANTO JEYA DAYALAN, N. EDISON AND S. PRAKASH
P.G. Department of Biotechnology, Udaya School of Engineering, KANYAKUMARI (T.N.) INDIA

(Accepted : February, 2009)

The oxygen mass transfer coefficient (K_La) often serves to compare the efficiency of bioreactors and their mixing devices as well as being an important scale up factor. In submerged fermentation, four methods are available to estimate the overall oxygen mass transfer coefficient. Each method provides a distinct estimation of the value of K_La. Dynamic gassing out method was used to obtain a more probable value of K_La during the fermentation in 5L fermentor with *Pichia pastoris*. The aeration and agitation were varied to study the effect on K_La. Results showed that the K_La value increased with increase in agitation and aeration. While working in 5L fermentor with *Pichia pastoris* the maximum K_La value of 441.36 was obtained when the agitator speed 300 RPM and aeration 1.5 vvm was maintained. This shows that the volumetric oxygen transfer rate varies in variation with aeration and agitation and not based on organism used. The optimum conditions of agitation and aeration to achieve maximum K_La for a given microorganism depends on its shear sensitive nature.

Key words : K_La, *Pichia pastoris*, Aeration, Agitation, Dissolved oxygen.

INTRODUCTION

The supply of oxygen is critical factor in all aerobic fermentations. An insufficient oxygen transfer leads to a decrease of microbial growth and product formation. In order to assess if particular equipment would be able to supply oxygen at a non-limiting rate, it is essential to have a good estimate of oxygen mass transfer coefficient (K_La). In submerged fermentation oxygen mass transfer coefficient serves to compare the efficiency of bioreactors and their mixing devices. It is also one of the important scale-up factors.

Hirose and Shibai's (1980) investigations of aminoacid biosynthesis by *Brevibacterium flavum* provide an excellent example of the effect of the dissolved oxygen concentration on the production of a range of closely related metabolites. These workers demonstrated the critical dissolved oxygen concentration for *B. flavum* to be 0.01 mg dm^{-3} and considered the extent of oxygen supply to the culture in terms of the degree of 'oxygen satisfaction', that is the respiratory rate of the culture expressed as a fraction of the maximum respiratory rate. Thus, a value of oxygen satisfaction below unity implied that the dissolved oxygen concentration was below the critical level. An example of the effect of dissolved oxygen on secondary metabolism is provided by Zhou *et al.* (1992) work on cephalosporin C synthesis by *Cephalosporium acremonium*. These workers demonstrated that the critical oxygen concentration for cephalosporin C synthesis during the production phase was 20% saturation. At dissolved oxygen concentrations below 20% cephalosporin C concentration decline and penicillin N increased. Bartholomew *et al.* (1950) represented the transfer of oxygen from air to the cell, during a fermentation, as occuring in a number of steps as transfer of oxygen from air bubble in to solution, then transfer of dissolved oxygen through fermentation media to the microbial cell, finally uptake of dissolved oxygen by the cell.

Many methods for determination of K_La in submerged fermentation have been performed with water and other model fluids, in order to mimic as closely as possible conditions encountered in fermentation systems. These investigations are very useful because conditions are well defined and can be rigorously controlled, and provide fairly good estimates of the oxygen mass transfer that can be used in design calculation. The determination of oxygen absorption from air to fermentation broth should, however, be assessed under actual operating conditions of fermentors since the rate of oxygen absorption into a culture medium can be greatly affected by presence of microorganisms, substrate, substances excreted by microorganisms and antifoam. K_La values in fermentors often differ substantially from values predicted for oxygen absorption in water.

In submerged fermentation, four methods are available to estimate the overall oxygen mass transfer coefficient: Sulphite oxidation method, Static gassing out method, Dynamic gassing out method and Oxygen balance