

**10C    HYPERBARIC BIOREACTORS: TOOLS FOR MASS  
02      TRANSFER AND CELL PHYSIOLOGY STUDIES**

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Laboratory research of microbial cultivation process is usually performed at atmospheric pressure, but in industrial bioreactors of several tens meters high, pressure and gas solubilities are function of the local position in the bioreactor, generally increasing by 1 bar for every 10 m increase in depth. Hyperbaric bioreactors are very useful tools to study increased pressure effects on cell physiology which has an important impact on bioprocess overall productivity. On the other hand, the increase of air pressure is useful to enhance oxygen mass transfer in aerobic cultures, mainly in high oxygen demand cultures. However, limits of pressure increase exists due to oxygen and carbon dioxide toxicity to microorganisms.

The effects of gas pressure rise on microbial cells strongly depend on the nature of gas, strain, mode of operation, etc. Hyperbaric bioreactors have been used to investigate the behaviour of bacteria and yeasts under hyperbaric stress. Besides metabolic and oxidative stress cell response to pressure, morphological changes have also been assessed through novel image analysis procedures.

*Saccharomyces cerevisiae* has been the main cellular model used to study the effects of air pressure in microorganisms. An adaptation cellular mechanism to hyperbaric air has been identified in fed-batch cultures of this yeast (Belo et al. 2003). This behaviour was also found in strains of *Kluyveromyces marxianus* exposed to sub-lethal air pressures and subsequently exposed to high oxygen pressures or chemical oxidants (Pinheiro et al, 2002). Also, a *Candida utilis* strain was able to grow under hyperbaric air and an enhancement of biomass productivity was achieved in high-cell-density cultures.

All of the yeast strains studied could cope with hyperbaric pressure of air and N<sub>2</sub>. The inhibitory effects to cell activity observed at air pressures above 10 bar were due to the oxygen effect and not to the total pressure.

The resistance to oxidative stress depends on the cell ability to induce their defensive antioxidant enzymes and the existence of limited antioxidant cell capacity is one of the reasons of oxygen toxicity. Thus, opposite effects are present in yeast aerobic cultivation, one is the oxygen availability and the other is the oxidative stress imposed. A model that takes into account the effect of oxygen toxicity on cell viability and growth was developed and successfully used to describe X-P-S evolution of *S. cerevisiae* cultures (Coutinho et al, 2004).

Similarly to oxygen, carbon dioxide have strong inhibitory effects on yeasts, resulting in viability loss, cell growth inactivation and morphological changes (Coelho et al, 2004).

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