## On-line estimation of biomass through pH control analysis in aerobic yeast fermentation systems

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Biomass determination is a basic parameter in fermentation processes. Therefore, simple and reliable on-line estimation procedures are highly desirable, particularly in fermentation processes using *Saccharomyces cerevisiae* [1,2], which are widely used in industry.

Experimental evidence of the existence of a direct relationship between proton production and growth has been presented in the past [3,4]. The determination of the equivalents of acid or base consumed by the culture per unit time has been a widely used parameter for on-line control processes [5,6]. Nevertheless, the majority of studies have been restricted to the establishment of empirical relations, while the physiological basis of the models proposed remained unclear.

Many studies on changes in medium pH associated with growth processes pointed to metabolic activity as the principal cause of medium proton exchange [3,7]. This fact was confirmed in a recent study in which the nitrogen assimilation pathway was certified as the main contributing pathway under aerobic conditions and respiratory metabolism [8].

The aim of this work is to verify the applicability of the model described above to those aerobic conditions in which respiro-fermentative metabolism is involved with production and consumption of ethanol. Experiments have been performed with dilution rates ranging from 0.11 to 0.38  $h^{-1}$ , using urea as the nitrogen source, in order to evaluate the contribution of the fermentative pathway to the total specific rate of proton production or consumption by the culture (qH<sup>+</sup>, mmol<sup>-1</sup>  $h^{-1}$  gbiom<sup>-1</sup>), determined as in Castrillo and Ugalde [8].

The experiments have shown that production or consumption of ethanol does not contribute significantly to the specific rate of proton production  $(qH^+)$ , thus extending the previously obtained relations [8] for all aerobic conditions in which other major acid/base contributions are not involved. This constitutes the fundamental metabolic basis by which many fermentation processes can be monitored through the accurate titration of the pH control reagent. Those relations may be usefully applied in fermentation control processes as formal rules, extensive for many different microorganisms displaying similar physiological patterns. Tests in batch and chemostat culture confirm the validity of  $qH^+$  as a formal control parameter in fermentations.

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