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## Optimisation of a continuous flash fermentation for butanol production using the response surface methodology

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### A B S T R A C T

Factorial design and response surface techniques were used in combination with mathematical modelling and computational simulation to optimise an innovative industrial bioprocess, the production of biobutanol employing the flash fermentation technology. A parametric analysis performed by means of a full factorial design at two levels determined the influence of operating variables on butanol yield and productivity. A second set of simulations were carried out based on the central composite rotatable design. This procedure generated simplified statistical models that describe butanol yield and productivity as functions of the significant operating variables. From these models, response surfaces were obtained and used to optimise the process. For a range of substrate concentration from 130 to 180 g/l, the optimum operating ranges ensure butanol productivity between 7.0 and 8.0 g/lh, butanol yield between 19 and 22%, substrate conversion above 90% and final butanol concentration around 25 g/l.

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### 1. Introduction

Optimisation through factorial design and response surface analysis is a common practice in biotechnology. Usually this technique is applied for the optimisation of culture conditions and for the determination of optimal values for processing parameters, such as pH, temperature, aeration and feeding rate, among others. Nowadays the use of this technique to establish optimal process designs for industrial-scale fermentations, as well as for real time process integration purposes, is increasing and demonstrating to be efficient, especially when accompanied by the use of mathematical modelling and computational simulation (Silva et al., 1999; Kalil et al., 2000; Costa et al., 2001). For this reason, this approach is employed in this work for the optimisation of an industrial-scale fermentation for butanol production.

Carrying out the butanol fermentation under optimised operating conditions is essential to run a biobutanol industry that can compete effectively with the current butanol derived from the petrochemical route, since the ABE fermentation, as normally the fermentation to produce butanol is called, is characterised by its low productivity. In this fermentation, acetone, butanol and ethanol (ABE) are produced in the ratio 3:6:1, with butanol being the major product. Product toxicity results in low butanol concentration in the reactor. In addition, the use of dilute sugar solution results in large process volumes. Mainly because of these problems and due to high costs related to the distillation of dilute product streams, the production of biobutanol on a commercial scale has been considered to be uneconomical (Ishizaki et al., 1999; Ezeji et al., 2007).

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## Nomenclature

$F_0$	feed broth flow rate ( $\text{m}^3/\text{h}$ )
$F$	fermentor outflow rate ( $\text{m}^3/\text{h}$ )
$F_c$	inlet flow rate of the flash tank ( $\text{m}^3/\text{h}$ )
$F_p$	permeate flow rate ( $\text{m}^3/\text{h}$ )
$F_{pu}$	fermentor purge flow rate ( $\text{m}^3/\text{h}$ )
$F_r$	liquid outlet flow rate of the flash tank ( $\text{m}^3/\text{h}$ )
$F_{re}$	return stream flow rate ( $\text{m}^3/\text{h}$ )
$F_v$	condensate vapour outlet flow rate of the flash tank ( $\text{m}^3/\text{h}$ )
$K_i$	equilibrium constant
$P_{\text{flash}}$	flash tank pressure (kPa)
$P_i^{\text{sat}}$	vapour pressure of component $i$ (kPa)
$P_0$	inlet product concentration (g/l)
$P_i$	fermentor product concentration (g/l)
$P_r$	product concentration in the liquid outlet flow of the flash tank (g/l)
$P_v$	product concentration in the vapour outlet flow of the flash tank (g/l)
$r_x$	rate of cell growth (g/lh)
$r_s$	rate of substrate utilisation (g/lh)
$r_{p_i}$	rate of products production (g/lh)
$S_0$	inlet substrate concentration (g/l)
$S$	fermentor substrate concentration (g/l)
$S_r$	substrate concentration in the liquid outlet flow of the flash tank (g/l)
$S_v$	substrate concentration in the vapour outlet flow of the flash tank (g/l)
$T_{\text{ferm}}$	fermentor temperature ( $^{\circ}\text{C}$ )
$T_{\text{flash}}$	flash tank temperature ( $^{\circ}\text{C}$ )
$V$	total volume of the system ( $\text{m}^3$ )
$x_i$	liquid molar fraction of component ( $i$ )
$X_0$	inlet biomass concentration (g/l)
$X$	fermentor biomass concentration (g/l)
$X_c$	biomass concentration in the inlet flow of the flash tank (g/l)
$X_p$	biomass concentration in the permeate (g/l)
$X_v$	biomass concentration in the vapour outlet flow of the flash tank (g/l)
$y_i$	vapour molar fraction of component $i$
$\gamma_i$	activity coefficient

During the past two decades a significant amount of research has been performed on the development of alternative technologies designed to remove the butanol continuously from the fermentation broth (e.g. adsorption, gas stripping, ionic liquids, liquid–liquid extraction, pervaporation, aqueous two-phase separation, supercritical extraction, perstraction, etc.) (Ezeji et al., 2007). These recovery techniques reduce the effect of product inhibition allowing an increase in the substrate concentration, which results in a reduction in the process streams, higher productivity and lower distillation costs (Groot et al., 1992).

In the process presented in this work, the continuous recovery of the butanol is carried out by the flash fermentation technology (Roffler et al., 1984; Silva et al., 1999; Costa et al., 2000, 2001; Costa and Maciel Filho, 2004; Atala, 2004; Mariano et al., 2008), in which the fermentor remains at atmospheric pressure and the broth is circulated to a vacuum chamber where butanol is continuously boiled off. A statistical methodology (factorial design) was applied to this process in order to

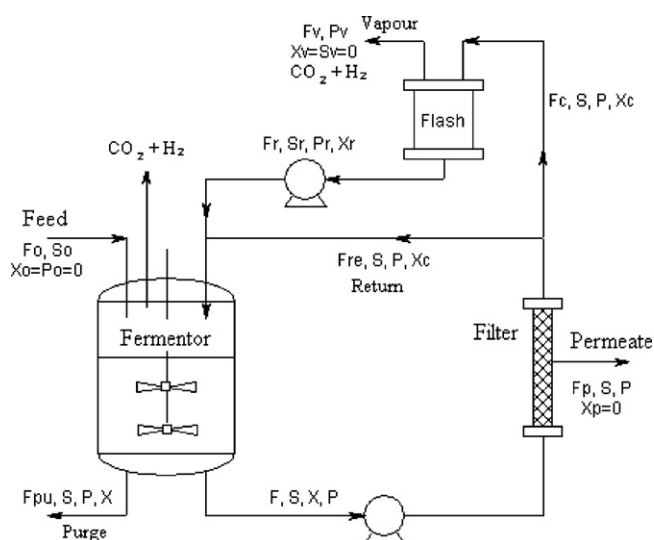


Fig. 1 – General scheme of the continuous flash fermentation process.

determine the most important operating variables for the optimisation of the process and the technique of surface response was used to find the best ranges of operating conditions that maximise butanol yield and productivity.

## 2. Process description and mathematical modelling

Fig. 1 depicts the flash fermentation process, which is a continuous fermentor connected to a cell retention system (filter) and an in-line product recovery equipment (flash tank). The broth is continuously circulated through the cell retention system in order to increase the biomass concentration in the fermentor. Product toxicity is reduced by partially recovering the solvents in the flash tank, which is placed in the recirculation line between the cell filter and the fermentor. Cell bleeding is carried out in the purge stream in order to avoid excessive cell growth.

Thus in the flash fermentation process there are three interconnected units, as follows: fermentor, cell retention system (tangential microfiltration) and vacuum vessel. The process starts as a conventional continuous fermentation until steady state is reached. Then, the flash tank separation system is turned on (i.e. vacuum is applied and pressure in the flash tank is reduced to 6.50 kPa), where a partial separation of the solvents and water mixture occurs. The liquid fraction ( $F_r$ ) returns to the fermentor and the vapour fraction ( $F_v$ ) after being condensed is combined with the purge ( $F_{pu}$ ) and permeate ( $F_p$ ) streams. These three streams ( $F_v$ ,  $F_p$ , and  $F_{pu}$ ) compose the final stream that is sent to distillation. The  $F_{re}$  stream (return) can be activated to regulate the inlet flow rate of the flash tank ( $F_c$ ).

The cell retention system allows the fermentor to be operated at high dilution rates without cell washout. The cells remain suspended in the liquid medium and a membrane is used as a means of preventing the cells from being removed with the out flow. For the mass balance, it is assumed that all cells are retained in the filter and solubilised compounds (substrate and products) freely pass through the membrane. Thus in the permeate, the cell concentration is equal to zero ( $X_p = 0$ ) and the concentrations of solubilised compounds ( $S$  and  $P$ ) are the same as in the fermentor.

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