

PRODUCTION OF ETHANOL BY COUPLING
FERMENTATION AND SOLVENT EXTRACTION

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SUMMARY

A new technology of fermentation is proposed. The inhibitor product is removed continuously by coupling fermentation and solvent extraction. Applied to ethanol fermentation this technology is suitable to any case where the terminal product is inhibitory.

The proposed technology uses both plug flow reactor and liquid-liquid extraction to achieve continuously the extractive fermentation of ethanol. The solvent used for liquid-liquid extraction is dodecanol. A new reactor was used. It is a column packed with a porous material. The fermentation broth is pulsed (a) to increase the interfacial area between the liquid medium and the dodecanol, and (b) to decrease the gas hold up.

Alcoholic fermentations were performed on glucose syrup at 35°C using *Saccharomyces cerevisiae*, with adsorbed cells as reference, with adsorbed cells and extractive fermentation. The results show that the fermentation is substantially improved. By this new method the ethanol productivity was multiplied by 5 and a solution of 407 g/l of glucose was totally fermented with a yeast which cannot normally transform more than 200 g/l glucose.

INTRODUCTION

Conventional ethanol fermentation is limited by the inhibitory effect of the product which decreases the rate of ethanol production and the cell biomass concentration giving a final alcohol concentration not more than between 10 to 15% V/V, from which the ethanol must be recovered by distillation. One way to reduce these limitations is to remove ethanol during its production, a process which we may call extractive fermentation.

In alcoholic fermentation, the vacuum fermentation process is one solution. Proposed first by Boeckeler (1948), described by Ramalingam and Finn (1977), Cysewski and Wilke (1977) the efficiency of this process is now established; however, the energy cost is a major disadvantage: Ghose and Tyagi (1979). Recently Maiorella and Wilke (1980) established that the vacuum fermentation process has an overall energy requirement for production of azeotropic ethanol of $8.352 \cdot 10^6$ J/m³ of ethanol. However, the high productivity in ethanol production is attractive.

It is well known that ethanol is extractible from water by liquid-liquid extraction: Hartline (1979). Alcohol dissolves readily in some liquids that do not mix with water. By exploiting this solubility difference, alcohol can be recovered by solvent extraction, but for extractive fermentation the non toxicity of solvent toward the cells must also be established. Recently, Goma et al. (1980),

Minier and Goma (1980) described dodecanol as solvent and proposed to use liquid-liquid extraction to remove ethanol produced during continuous fermentation with immobilized cells, while Hernandez Mena and al. (1980) established the process feasibility of continuous extraction by dibutyl phthalate in batch or fed batch culture. The aim of this paper is to describe a new technology of fermentation which combines both liquid-liquid extraction and fermentation and to show the improvement which can result.

MATERIAL AND METHODS

The yeast used was *Saccharomyces cerevisiae* UG5 described in a previous work : Strehaiano and al., 1978. The medium contained (gl^{-1}) KH_2PO_4 , 5 ; $(\text{NH}_4)_2\text{SO}_4$; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.4 ; yeast extract, 1 ; glucose, variable ; tap water to 1 liter pH adjusted to 3,75 with H_3PO_4 after sterilization. Biomass concentration was determined by filtration (Millipore 0.45 μ) weighing after 12 h at 0.2 B, 60°C. Ethanol was measured by G.L.C. (Perkin Elmer F 11) with n-butanol as internal standard, glucose by dinitrosalicylate method and viability by coloration with Ponceau red.

The "dodecanol" used for extractive fermentation is sold by Prolabo, Paris, ref. 20 840 292. This commercial product was found to be a mixture of n-dodecanol (60%), n-tetradecanol (40%), and n-decanol (trace).

RESULTS

1) Choice of a technology of extractive fermentation

The technology of extractive fermentation coupling both liquid-liquid extraction and fermentation must have : (a) compatibility with a continuous process (b) plug flow hydrodynamics for the fermentation phase and the solvent.

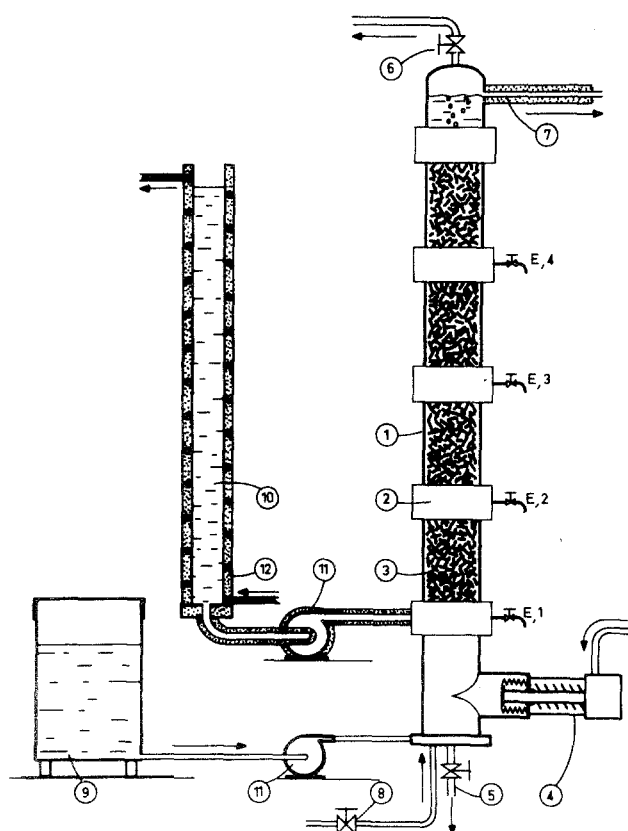


Fig. 1 : Device for continuous extractive fermentation coupling liquid-liquid extraction and ethanol production by immobilized cells.
 1. reactor ($H = 1.7 \text{ m}$, $\phi_{\text{int}} = 0.1 \text{ m}$), 2. heat exchange, 3. packing, 4. pulse generator (EIVS), 5, 6, 7. outlets, 8. air input (occasional), 9. aqueous medium reservoir, 10. dodecanol reservoir, 11. metering pumps 12. heater (to keep dodecanol molten).

We developed for SCP production a new type of continuous fermentor which features an agitation-aeration system hand on pulsed flow across perforated plates : Serieys and al. (1978). This device was packed with porous brick in order to increase the biomass concentration in the reactor and to improve the liquid-liquid extraction by dodecanol. It is fed both by dodecanol and by fermentation medium. The device used for extractive fermentation is shown in figure 1 and has been run continuously during 1,5 years:

2) Improvement of alcoholic fermentation by extractive fermentation

Continuous cultures were performed with immobilized cells with or without extractive fermentation. The results were obtained in extreme conditions for *Saccharomyces cerevisiae*.

Three series of experiments are reported one of reference without dodecanol, the other, with only a tiny amount of dodecanol, for the purpose of evaluating the additive effect of dodecanol without any removal of ethanol, (surfactant effect of dodecanol, increase of the yeast clumping). The third series of experiments are extractive fermentation.

Results obtained are summarized in table I ; obviously, alcoholic fermentation is improved by extractive fermentation. The residual sugar drops from 162 gl^{-1} to 2 gl^{-1} while the productivity is multiplied by 4 with input glucose at 260 gl^{-1} and by 5 at 317 gl^{-1} .

Table I : Continuous alcoholic fermentation with immobilized cells without dodecanol addition (control), with very small addition of dodecanol (experiment 1), with extractive fermentation (experiments 2, 3, 4).

| Experiments | Control | 1 | 2 | 3 | Control | 4 |
|--|---------|--------|--------|--------|---------|-------|
| Parameters | | | | | | |
| Dodecanol feed (lh^{-1}) | 0 | 0,0285 | 1 | 2,55 | 0 | 1 |
| Medium feed (lh^{-1}) | 0,069 | 0,0667 | 0,0728 | 0,0698 | 0,068 | 0,068 |
| Glucose input (gl^{-1}) | 260 | 263 | 260 | 263 | 315 | 317 |
| Glucose output (gl^{-1}) | 162 | 165,9 | 2 | 1,8 | 242 | 2,9 |
| Ethanol conc. in aqueous phase (gl^{-1}) | 38,1 | 39,8 | 21,2 | 9,4 | 28,9 | 23,7 |
| Ethanol conc. in dodecanol phase (gl^{-1}) | | 14,9 | 8,38 | 3,35 | | 9,1 |
| Total ethanol productivity ($\text{gl}^{-1}\text{h}^{-1}$) | 0,24 | 0,28 | 0,85 | 0,90 | 0,18 | 0,95 |
| Ethanol yield constant (g.g^{-1}) | 0,4 | 0,47 | 0,51 | 0,5 | 0,4 | 0,5 |

DISCUSSION CONCLUSIONS

The demonstration of the process feasibility for continuous extraction of ethanol from a continuous fermentation system is established and indicates the efficiency of this new technique. In addition, the yield obtained is close to the theoretical maximum yield. This is not a casual observation but we cannot explain it.

Extractive fermentation reduces the effect of product inhibition and in

effect the sugar tolerance of this strain is highly increased. Our most recent results indicate complete fermentation of 407 g/l glucose solutions.

One of the most important problems in the ethanol fermentation is to lower the recovery costs. By the utilization of a membrane technique for ethanol separation from the dodecanol and by recycling the mineral medium it should be possible to decrease the energy cost of ethanol recovery and though more work is necessary, the possibility of developing an isothermal ethanol recovery process appears to be realistic.

Extractive fermentation using liquid-liquid extraction should be applicable to all fermentations where the fermentation product can cause inhibition and/or repression of its own synthesis or can affect microbial growth.

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