THE EFFECT OF CO ON GROWTH AND PRODUCT FORMATION IN BATCH CULTURES OF CLOSTRIDIUM ACETOBUTYLICUM

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Summary. Carbon monoxide sparged in batch fermentations of C. acetobutylicum inhibits the production of H₂ by the hydrogenase and enhances the production of solvents by making available larger amounts of NAD(P)H₂ to the cells. CO also inhibits biomass growth and acid formation. Its effect is most pronounced under fermentation conditions of excess carbon- and nitrogen-source supply.

INTRODUCTION

The elucidation of the mechanisms which trigger and control solvent production in fermentations of butyric-acid bacteria is a problem of both fundamental and practical importance, and has recently become the subject of vigorous research activity (Kim et al., 1984; Papoutsakis, 1983). We have recently postulated that solvent production is predominantly regulated by the availability and demand of biosynthetic (ATP) and reduction (NAD(P)H₂, FdH₂) energies (Papoutsakis, 1983; Roos et al., 1985). We have shown that conditions of reduced nitrogen-source availability in batch and continuous cultures of C. acetobutylicum (whereby glucose and thus ATP were in excess supply) induce solvent formation (Roos et al., 1985). On the contrary, under glucose-limited conditions, acids are exclusively produced (Roos et al., 1985). The hypothesis that solvent production is controlled by the demand and availability of ATP appears indeed valid. We have also calculated the values of NfF, defined as the amount of NAD reduced by FdH₂ (via the NADH: ferredoxin oxidoreductase) as a function of time for a number of fermentations, using the fermentation equation which has been derived and validated earlier (Papoutsakis, 1984). We found that in all acid fermentations, NfF is negative (i.e. Fd is reduced by NADH₂) throughout the fermentation, while large rates of increase in NfF values and positive NfF values precede and accompany solvent production only (Roos et al., 1985). Thus, the rate of change and values of NfF can predict solvent formation, indicating that, perhaps, an excess availability of reduction energy (which is otherwise released as H₂) would induce solvent formation. In order to examine this possibility, we sought a means of making available larger amounts of reduction energy to the cells, to investigate how this would affect the distribution of products. This we accomplished by blocking H₂ production using CO, which is a known inhibitor of the hydrogenase (Gray and Gest, 1965; Mortenson and Chen, 1975). Here we report some of our results with batch cultures under conditions of both excess and limited nitrogen-source supply. A detailed report on the effect of CO on the microbial metabolism in continuous cultures will be forthcoming. The idea of employing CO to probe the metabolism of C. acetobutylicum has been investigated, apparently independently of and simultaneously with our own efforts, by Kim et al (1984) [Our first conclusive experiments were performed in the summer of 1983 and were reported in a proposal submitted to the National Science..."
FIGURE 1