



Fig. 2. Effect of (a) substrate concentration and (b) time course of simultaneous saccharification and fermentation of alkali-treated newspaper (5%) for ethanol production.

that the pretreatments make the substrate more susceptible to cellulase attack and hence increased saccharification. Our findings on saccharification are in accordance with the results of Andren *et al.* (1976) who noted 50% conversion of waste-paper cellulosic material using *T. reesei* cellulase. Mandels *et al.* (1974) noted 55.6% saccharification of milled newsprint after 48 h incubation.

Figure 2 depicts a gradual increase in ethanol production with increasing substrate from 1 to 5%. After 96 h fermentation the ethanol remained constant up to 120 h. This could be attributed to either exhaustion of nutrients and/or an inhibitory effect of ethanol. Ghose *et al.* (1982) suggested that *T. reesei* QM 9414 cellulase is inhibited uncompetitively by ethanol. Ooshima *et al.* (1985) reported that less than 4M ethanol did not affect the enzymatic activities of β -glucosidase and *endo*-glucanase.

ACKNOWLEDGEMENT

This paper was prepared through the Director, Experiment Station, G.B. Pant University of

Agriculture and Technology, Pantnagar, India as research paper No. 6407.

REFERENCES

- Andren, R. K., Erickson, R. J. & Medeiros, J. E. (1976). Cellulosic substrate for enzymatic saccharification. *Biotechnol. Bioeng. Symp.*, **6**, 177-203.
- Chavadej, S., Mishra, C., Rao, M., Seeta, R., Srinivasan, M. C. & Jagannathan, V. (1980). Utilization of cellulosic waste for energy production. *Regional J. Energy*, **2**, 23-9.
- Deschamps, A. M., Gillic, J. P. & Ubeault, J. M. (1981). Direct delignification of untreated bark chips with mixed culture of bacteria. *Eur. J. Appl. Microbiol. Biotechnol.*, **13**, 222-5.
- Franzidis, J. P. & Porteus, A. (1981). In *Fuels from Biomass and Wastes*, ed. D. L. Klass & G. H. Emert. Ann Arbor Science Publisher, p. 267.
- Furusaki, S., Nobuhiro, A. & Kiyochika, H. (1986). Saccharification of sweet sorghum bagasse by cellulase. *J. Ferment. Technol.*, **63**, 523-8.
- Ghose, P., Pamment, N. B. & Martin, R. B. (1982). Simultaneous saccharification and fermentation of cellulose: effect of β -D-glucosidase activity and ethanol inhibition of cellulases. *Enz. Microbiol. Technol.*, **4**, 425-30.
- Graham, E. R. (1948). Determination of soil organic matter by means of photoelectric colorimeter. *Soil Sci.*, **65**, 181-3.
- Johnson, D. B., More, W. E. & Zank, L. C. (1961). The spectrophotometric determination of lignin in small wood samples. *Tappi*, **44**, 793-5.
- Mandels, M., Hontz, L. & Nystrom, J. (1974). Enzymatic hydrolysis of waste cellulose. *Biotechnol. Bioeng.*, **16**, 1471-93.
- Miller, G. L. (1959). Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal. Chem.*, **31**, 426-8.
- Neish, A. C. (1971). Analytical methods for bacterial fermentation. In *Methods in Microbiology*, ed. J. R. Norris & D. W. Ribbons, Vol. 6, Academic Press, New York, pp. 53-215.
- Ooshima, H., Ishitani, Y. & Harano, Y. (1985). Simultaneous saccharification and fermentation of cellulose: effect of ethanol on enzymatic saccharification of cellulose. *Biotechnol. Bioeng.*, **27**, 389-97.
- Pamment, N., Robinson, C. W. & Young, M. M. (1979). Pulp and paper mill solid waste as substrates for single cell protein production. *Biotechnol. Bioeng.*, **21**, 561-73.
- Sternberg, D. (1976). Production of cellulase by *Trichoderma*. *Biotechnol. Bioeng. Symp.*, **6**, 35-53.
- Updegraff, D. M. (1969). Semi-microdetermination of cellulose in biological materials. *Anal. Biochem.*, **32**, 420-4.
- Ward, G. M., Muscato, T. V., Hill, D. A. & Hausen, R. W. (1978). Chemical composition of feedlot manure. *J. Environ. Quality*, **7**, 159-64.
- Warzywoda, M., Ferre, V. & Pourquie, J. (1983). Development of culture medium for large scale production of cellulolytic enzymes by *Trichoderma reesei*. *Biotechnol. Bioeng.*, **25**, 3005-10.