BIOCOMPATIBILITY ASSESSMENT OF PERFLUOROCHEMICAL OILS IN MICROBIAL AND PLANT CELL CULTURES

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SUMMARY

The effects of various perfluorochemical (PFC) oils on growth and structure of microbial and plant cell cultures have been studied. Growth of microbial cells was unaffected by culture with PFCs and no obvious deleterious effects on cell structure or alterations in polypeptide profiles of cell extracts were observed. Incubation of S. dulcamara cells with FDC oil produced an increase in culture growth rate, as measured by changes in wet and dry cell weights, probably by providing protection against hydrodynamic damage.

INTRODUCTION

PFCs have been proposed as supplements in prokaryotic and eukaryotic cell cultures to improve gas supply and provide protection against mechanical damage produced by conventional aeration methods (Mattiasson and Adlercreutz, 1987; King et al., 1989). PFCs are chemically inert and have the capacity to dissolve substantial volumes of respiratory gases (Riess and Le Blanc, 1982, 1988). In addition, they are immiscible with aqueous systems, making them recoverable and hence, potentially recyclable. However, the biocompatibility of PFC oils in cell culture systems has not yet been studied in detail and this is especially the case with plant cells. We have therefore investigated the effects of different PFC oils on growth and structure of Escherichia coli, Saccharomyces cerevisiae and Solanum dulcamara cells in suspension culture. Part of this investigation involved electrophoretic analysis of yeast protein profiles following culture with PFCs.

MATERIALS AND METHODS

Organisms; media and growth

Experimental cultures of E. coli (HB101 containing plasmid pBR322) and S. cerevisiae (X 2180 1B) were grown at either 37°C (E. coli) or 30°C (yeast) as described in detail previously (King et al., 1988a). Callus cultures of S. dulcamara were initiated from 15-20 day-old plants by placing sterilized [15% v/v Domestos (Lever, Kingston) for 15 min followed by washing 3 times in sterile water] leaf segments.

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in basal medium (Murashige and Skoog, 1962) supplemented with 2.0 mg alpha-naphthaleneacetic acid/L and 0.5 mg 6-benzylaminopurine/L. Plant cell cultures were maintained at 25 ± 2°C in continuous fluorescent light (1.0 W m⁻²) and agitated at 70 rpm; sub-cultures and test cultures were prepared every 7 days by adding 10 ml of culture to 40 ml fresh medium.

The following PFC oils were obtained from RTZ Chemicals, Avonmouth: perfluorodecalin (FDC), perfluorotripropylamine (FTPA), and perfluorotributylamine (FTBA)(Figure 1). Microbial cell cultures (10 ml) containing 5-30% (v/v) PFC were grown for up to 450 min; culture growth was determined by measurement of changes in optical density (600 nm) and viable cell counts using conventional serial-dilution and agar plating (King et al., 1988a). Plant cell cultures containing 30% (v/v) PFC (sterilized by autoclaving at 121°C for 20 min) were grown for up to 9 days; culture growth was determined by measurement of both fresh and dry cell weights.

![Chemical structure of PFCs.](image)

**Figure 1.** Chemical structure of PFCs.

**Statistical analysis**

In the case of microbial cell studies, statistical analyses were performed according to the methods of Snedecor and Cochran (1980). Means and standard errors (SEM) have been used throughout and statistical significance between mean values was assessed using a conventional Student’s t test. For plant cell studies, two-way analysis of variance without replication was performed according to the methods of Sokal and Rohlf (1981). A probability of P < 0.05 was considered significant.