METABOLIC EFFECTS OF 2-PHENYLETHANOL, 1,1-DIMETHYLPHENYLETHANOL, PERIODATE AND SUCCINIC ANHYDRIDE ON INTACT YEAST CELLS AND ON FERMENTING PREPARATIONS FROM LYOPHILIZED YEAST

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1. Introduction

Preceding reports dealt with the action of 2-phenylethanol (PEA) 1,1-dimethylphenylethanol (DMPEA), succinic anhydride, and periodate on membrane permeability and metabolism of ascites tumour cells [1,2] and erythrocytes [3,4]. It was concluded that the metabolic effects brought about by these compounds must be attributed, directly or indirectly, to membrane alterations. To support this assumption, comparative metabolic studies with intact red cells and hemolysates under the influence of the agents were carried out [3,4].

We feel that further evidence might be provided by the relatively simple device of comparing metabolic alterations of intact yeast cells with those of organelle-free preparations which still retain the entire fermenting system. More details on the properties of the agents and references may be found in [1], [2] and [4].

2. Materials and methods

Brewer's yeast was obtained from Sinner Brauerei AG, Karlsruhe, and prepared for metabolic assay by suspension (1:200 w/v) in phosphate buffer, adjusted to pH 6.

Organelle-free suspensions were prepared by freeze-drying of crumbed yeast (20 hr), dissolution in distilled water (1:5 w/v) and centrifugation. The organelle-free supernatant was further diluted (1:1 v/v) in phosphate buffer, and adjusted to the same pH and ionic strength as with intact cells.

Respiration and fermentation of glucose was determined by classical Warburg manometry. Anaerobiosis was produced by gassing with pure nitrogen.

3. Results and discussion

The effects of the amphipathic alcohols PEA and DMPEA, and the specific reagents periodate and succinic anhydride on yeast fermentation, as shown in figs. 1–3, may well be predicted from previous studies on ascites tumour cells and red cells [1–4]. The differences between metabolic rates of intact cells on the one hand, and lyophilized yeast preparations on the other, is especially obvious with PEA, DMPEA (fig. 1), and succinic anhydride (fig. 2), and clearly

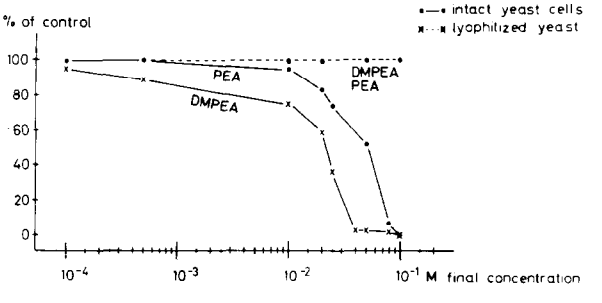

Fig. 1. Anaerobic CO_2-production of intact yeast cells (———) and lyophilized yeast preparations (– – – –) after treatment with 2-phenylethanol (PEA) and 1, 1-dimethylphenylethanol (DMPEA). Incubation 15 min at 37°C; pH 6.0; addition of 30 mM glucose after incubation. n = 12

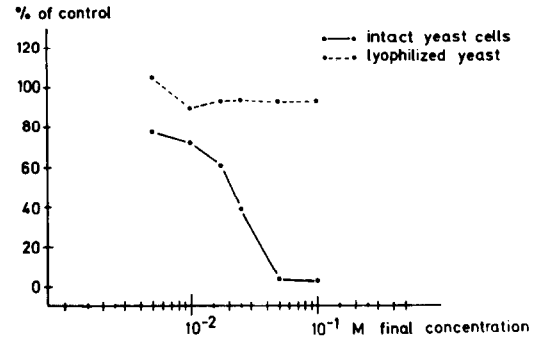

Fig. 2. Anaerobic CO_2-production of intact yeast cells (———) and lyophilized yeast preparations (– – – – –) after treatment with succinic anhydride. Incubation 15 min at 37°C, pH 6.0; addition of 30 mM glucose after incubation. $n = 12$

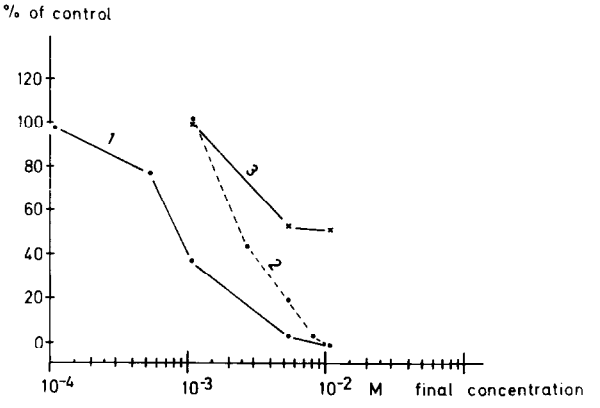

Fig. 3. Respiration of intact yeast cells (1), anaerobic CO_2-production of intact cells (2) and of lyophilized yeast preparations (3) after treatment with sodium periodate. Incubation 15 min at 37°C, pH 6.0; addition of 30 mM glucose after incubation. $n = 12$

points to an involvement of the cell membrane in metabolic inhibition of intact cells. It was to be expected, too, that DMPEA has a stronger effect than the parent compound. An impairment of yeast cell growth and an alteration of membrane permeability by PEA have been described by Burns [5,6]. Yet, this author could not confine growth inhibition solely to membrane alterations. As far as the effects on glucose fermentation are concerned, our results imply an exclusive involvement of the cell membrane. In fig. 3 respiratory activity of intact cells is included in order to demonstrate that with IO_4^- additional intracellular events may be involved. The finding that periodate affects cell-free preparations, too (although only at higher concentrations), must be attributed to direct interactions of the compound with critical nucleotides. The results with yeast add some more evidence to our conclusion from previous studies [1–4]: damage to intracellular systems plays no or only a subordinate role in metabolic alteration of intact cells by the four compounds, and the effects rather have to be attributed to membrane changes. Possible mechanisms have been proposed previously [1–4].

Acknowledgement

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