

Yeast Surface Potential Probed by 9-Aminoacridine

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According to Searle and Barber (1) 9-aminoacridine may be used to probe the electrical double layer associated with negatively charged biological membranes. They postulated that 9-AA binding to these membranes is purely based on an electrostatic interaction of the dye with the membrane.

We have now examined the applicability of this dye to probe the yeast surface potential. We studied the binding of the dye to the yeast *Saccharomyces cerevisiae*, Delft II, as a function of the dye concentration and cell density, and of the pH and ionic strength of the medium. Only in a limited concentration range of the dye, depending on the cell density, a linear relationship between dye binding and free dye concentration is found. Under these limited conditions, addition of salts or a reduction in pH of the cell suspension appreciably decreases dye binding to the yeast cells. The effect of salt addition appears to be primarily that of shielding the fixed negative charges, whereas the pH effect results from protonation of these groups. This concept is supported by the fact that the effectiveness of salts in reducing dye binding is as predicted by classical double layer theory, governed mainly by the charge carried by the cation with an order of effectiveness $C^{3+} > C^{2+} > C^{1+}$. Relatively small differences in the effectiveness of divalent cations in reducing dye binding are observed probably due to differential binding of these cations to the fixed charges.

The present results suggest that, at least qualitatively, 9-AA can be used to probe the yeast surface potential. The finding that dye binding to electrophoretically distinct yeast strains decreases in the same order as the zeta potential of the yeast cell does, supports that notion. We will show, however, that besides a simple electrostatic interaction of 9-AA with the yeast membrane surface probably other ones are involved. The applicability of 9-AA to quantize the yeast surface potential is therefore questionable. In addition, it is not completely excluded that part of the dye binding occurs to negative charges in the yeast cell wall.

1. Searle, G.F.W. and Barber, J. (1978) *Biochim. Biophys. Acta* 502, 309-320.

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