A NOVEL TECHNIQUE FOR STUDYING THE EFFECT OF ANTI-BACTERIAL AGENTS ON STAPHYLOCOCCUS AUREUS

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This communication reports the development of a new, physiologically satisfactory method for the simultaneous determination of ΔpH & $\Delta\Psi$, the osmotic and electrical components respectively of the bacterial proton motive force (Δp) (Mitchell 1966), and its application in studying the effect of a potent uncoupler, carbonyl cyanide m-chlorophenylhydrazone (CCCP) on Δp.

The addition of glucose to starved, potassium depleted S. aureus (4×10^9 cells ml^{-1}) results in the immediate uptake of potassium and the production of acetic acid, a by-product of glucose metabolism. Potassium uptake is believed to occur in response to $\Delta\Psi$ and by applying a modification of the Nernst equation to the uptake data (Denyer 1979) it was possible to calculate $\Delta\Psi$. In addition, since acetic acid will act as a freely permeable acid (Rottenberg 1975), the distribution ratio of the anion across the membrane at equilibrium was used to determine ΔpH according to the equation of Rottenberg (1975).

The results obtained can be compared with those determined by other more conventional methods of non-physiological origin (reviewed by Rottenberg 1975).

Table Comparison of values for ΔpH and $\Delta\Psi$ in S. aureus at pH 7.0.

Component	Technique used for determination				
	Acetate	Potassium uptake	DMO	DDA^+	$TMPB^+$
$\Delta\Psi$ (mv)	-	-148	-	-137	-148
ΔpH	0.82	-	1.00	-	-

The addition of 50 µM CCCP results in the partial collapse of ΔpH while a combination of CCCP and valinomycin (1 µg ml^{-1}) produces a marked decrease in both ΔpH and $\Delta\Psi$.

The advantage of this new technique lies in its speed, physiological acceptability (acetate is a normal metabolite) and its ability to examine simultaneously changes in both components of Δp. Many preservatives are believed to act in a similar way to CCCP and this technique may provide a useful method for screening for potential preservative action.

DMO - 5,5-Dimethyl-2,4-oxazolidinedione
DDA^+ - Dibenzyldimethylammonium cation
$TMPB^+$ - Triphenylmethylphosphonium cation

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