Description and Mechanisms of Bacterial Growth Responses to Water Activity and Compatible Solutes

by

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DECLARATION

I declare that this thesis contains no material which has been accepted for a degree or diploma by the University of Tasmania or any other institution, and to the best of my knowledge and belief contains no copy or paraphrase of material previously published or written by another person except where due acknowledgment is made in the text of the thesis.

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19/2/1998

ABSTRACT

Bacterial growth is inhibited by unfavourably low water activity conditions, however, the inhibition is partly alleviated by exogenous provision of compatible solutes. As compatible solutes exist in all food systems, describing and understanding this bacterial growth response is useful for food microbiologists concerned with limiting microbial growth on foods using water activity stress.

In the first application of predictive microbiology techniques to compatible solute growth responses, the four parameter square root model (Ratkowsky *et al.*, 1983) successfully described growth rate data collected for *Escherichia coli*, *Paracoccus halodenitrificans* and *Halomonas elongata*. The value of the parameter T_{min} was independent of the exogenous provision of compatible solutes, but $a_{w\ min}$ values and the observed minimum temperature for growth were lower where compatible solutes were provided.

Despite their accurate description of growth rate data, empirical square root models do not improve mechanistic understanding. A mechanistic explanation for the bacterial growth response to water activity and compatible solutes was examined using a substrate-limited batch culture technique, developed to measure cell yield. The cell yield of *E. coli* did not vary significantly with extracellular water activity or compatible solutes, except at water activity values close to the growth/no growth interface, indicating that water activity challenge is not an energetic burden for bacterial cells. Therefore, energetic limitation of growth was eliminated as a possible mechanistic explanation. Cell yield was also independent of incubation temperature, over most of the biokinetic range. The cell yield responses with water activity and temperature conditions were similar and consistent with a mechanistic, thermodynamic model (McMeekin *et al.*, 1993; Ross, 1993) , thus the influence of water activity on microbial growth may be explained in terms of the thermodynamics of protein folding. This mechanism is consistent with contemporary models for the effect of compatible solutes on water structure close to the surface of macromolecules (Wiggins, 1990).

Examination of novel and published data using the thermodynamic model revealed a possible mechanism for the temperature and water activity limits for microbial growth. Large increases in the activation energy, close to the boundary between growth permissible and non-growth conditions, suggest a possible universal limiting activation energy for bacterial growth. This finding may provide a mechanistic basis for, currently empirical, growth/no growth models.

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4. ACTIVATION ENERGY