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Edited by

R S KAHLON

Professor & Head,
Department of Microbiology
Punjab Agricultural University, Ludhiana

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GROWTH OF *HALOBACTERIUM* STRAIN R1 ON SODIUM BENZOATE

R. Aguair and I. Furtado*
Department of Microbiology, Goa University,Taleigao Plateau, Goa - 403 203.

Halobacterium strain R1, a halophilic archaebacterium, growing in 25% NaCl crude salt utilizes sodium benzoate as sole source of carbon and energy. Growth is accompanied with formation of three products.

Halobacterium strain R1, isolated from a salt pan (1) grows in 25% crude salt synthetic medium (2) containing 0.2% sugar. The present study reports the growth of this halophilic Archaebacterium on sodium benzoate as sole source of carbon.

Halobacterium strain R1 was grown and maintained at RT, on 0.2% glucose mineral salts medium agar containing per litre; 200g NaCl, 13g, $MgCl_2$. $6H_2O$; 20g, $MgSO_4$; $7H_2O$; 1g, $CaCl_2$, $2H_2O$; 4g, KCl; 0.2g, $NaHCO_3$; 0.2g, NH_4Cl; 0.005g, $FeCl_3$, $6H_2O$; 0.5g, KH_2PO_4; adjusted to pH7 with KOH (2) for growth of culture. 5 ml of a two day old 0.2% glucose mineral broth was inoculated into 250ml Erlenmeyer flask containing 100 ml mineral medium and 0.2% sodium benzoate and incubated at RT, on an Emenvee shaker at 150 rpm. The increase in turbidity was monitored at 480 nm using a spectrophotometer.

Transformation of sodium benzoate was analyzed by acidifying the cell free culture broth and extracting it with ether (40-80°C). The concentrate thus obtained was separated by thin layer chromatography (TLC), developed in benzene : methanol : acetic acid (45:10: 1v/v) and visualized with iodine vapours.

Halobacterium strain R1 grew in mineral salts medium containing 0.1-0.6% of sodium benzoate. Optimum growth was with 0.1% sodium benzoate with a lag of one day followed by a consistent log phase of 2-5 day that reached a stationary phase by 6 days (Fig.1). The culture broth contained three products with Rf. of 0.74, 0.61 and 0.34 more polar than sodium benzoate. These were designated as B_1, B_2, B_3 respectively. The substrate disappeared completely by the fifth day and the product B_3 could not be detected on the sixth day of the growth.

The nutritionally fastidious extreme halophiles are now known to grow in defined media with sugars (3-5). As yet there are no reports of extreme halophiles growing in 25% crude salt or NaCl using sodium benzoate as sole source of carbon and energy. This ability of *Halobacterium strain R1* is of interest as sodium benzoate is a widely used food preservative (6), alongwith high concentration of sodium chloride (brine) and that sodium benzoate is a model cyclic organic compound.

* Author to whom all correspondence should be addressed.

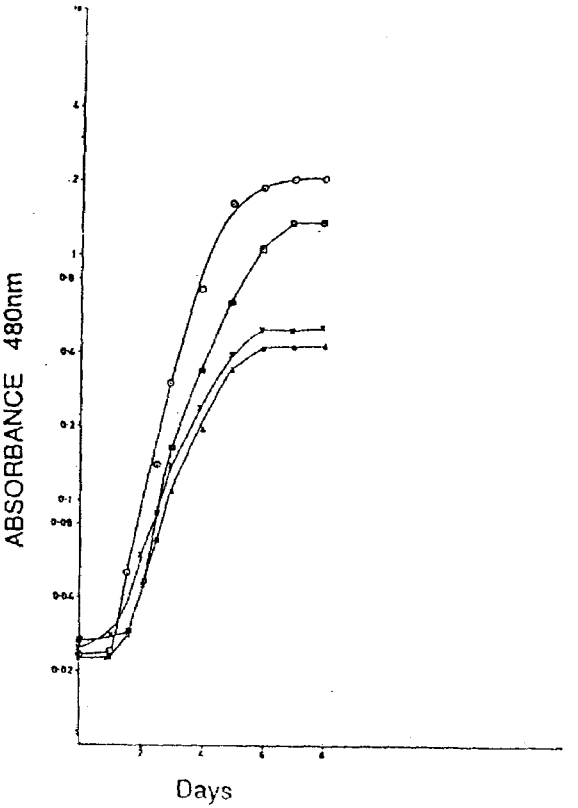

Fig. 1: Growth of halobacterium strain R1 on sodium benzoate.
0-0, 0.1%;□-□ 0.2%; x-x, 0.4%,Δ-Δ, 0.6%

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