

### A paraffin-oxidizing pseudomonad

A pseudomonad enriched from a Dublin soil in a mineral salts-paraffin medium has been found rapidly to oxidize hexadecane. Other alkanes of longer or shorter chain length were not oxidized without lag by either paraffin- or hexadecane-grown cells. Quantitative manometric observations showed that hexadecane was incompletely oxidized, and that a definite "break" in the oxidation rate corresponded to the consumption of 0.51 mole  $O_2$ /mole of substrate, which in turn implied production of cetyl alcohol as a major metabolic intermediate. Titration of the supernatant medium from cultures in early growth on mineral salts-paraffin medium indicated negligible acid production, but a faint dichromate reaction suggested that an alcohol had accumulated. In older cultures esters were detected but not identified.

Organisms in the late exponential growth phase in a hexadecane-mineral salts medium with little added nitrogen (20  $\mu$ g/ml ammonium nitrogen) were harvested, and the ethanol-ether-soluble fraction extracted from the air-dried cells. This fraction which accounted for 20 % of the dry weight was resolved to saponifiable and non-saponifiable lipids, and from the latter cetyl alcohol was isolated by crystallization from ethanol as flat plates, m.p. 49° which was not depressed by mixture with authentic cetyl alcohol. On nitrogen-rich medium (100  $\mu$ g/ml ammonium nitrogen) the cell yield was increased but total lipids were reduced to only 4 % of the dry weight, and no cetyl alcohol was recovered.

In contrast to the manometric behaviour of cells grown with ample nitrogen and hexadecane as carbon and energy source, cells grown on nitrogen-deficient medium with hexadecane as carbon source consumed a maximum of 0.5 mole oxygen/mole hexadecane; however, addition of ammonium nitrogen (100  $\mu$ g) to these nitrogen-deficient cells in the respirometer allowed further oxidation of the hexadecane. This further oxidation was sensitive to chloramphenicol, dinitrophenol, and azide, suggesting that it involved synthesis of a protein, probably an enzyme acting on cetyl alcohol as substrate.

This system provides an example of the synthesis of a series of simultaneously induced enzymes<sup>1</sup> the formation of one of which is influenced not only by the accumulation of its substrate, but also by the nitrogen level of the cells<sup>2</sup>.

Publication of these data, obtained in 1957 in the Department of Bacteriology, Trinity College, Dublin, has been unavoidably delayed and is now made since it provides independent confirmation, using different techniques, of the recent elegant studies of KALLIO and co-workers<sup>3</sup>.

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