SECTION H, MICROBIOLOGY

Pathway of Tryptophan Dissimilation[1]

NORMAN N. DURHAM and JERRY R. MARTIN

Department of Bacteriology, Oklahoma State University, Stillwater

The discovery of tryptophan in 1902 identified this amino acid as one of the more important metabolites associated with cellular growth and reproduction. Consequently, tryptophan metabolism has been studied in several biological systems. The microbial degradation of tryptophan by certain members of the genus *Pseudomonas* was elucidated by Stanier and Hayaishi (1951) using sequential induction. They found that these organisms utilized tryptophan by either the aromatic (anthranilic acid) or the quinolinic pathway (kynurenic acid).

A *Flavobacterium* sp. dissimilates both the D- and L-isomers of tryptophan via the aromatic pathway (Martin and Durham, 1961). This paper describes the ultimate fate of certain carbon atoms of the tryptophan molecule.

MATERIALS AND METHODS

All reactions were conducted in double side arm Warburg vessels. Isotopically labeled substrates were placed in one side arm and 0.2 ml of 2N HCl in the second side arm. A standardized cell suspension was pipetted into the main chamber and 0.2 ml of 20% KOH placed in the center well. The vessel was equilibrated to temperature and the reaction initiated by adding the substrate to the cell suspension. After the reaction had proceeded to completion, the acid was added to release the CO_2 absorbed by the buffer.

Samples of the cell suspension and KOH were analyzed for C^{14} by the wet combustion method and the radioactivity of the carbon dioxide determined with a vibrating reed electrometer.

RESULTS

Experiments using C^{14} labeled DL-tryptophan, anthranilic acid, and formic acid were conducted in an attempt to determine the ultimate fate of the carbon atoms in the tryptophan molecule. Carboxyl-C^{14}-anthranilic acid, 7A-C^{14}-DL-tryptophan, 1-C^{14}-anthranilic acid, carboxyl-C^{14}DL-alanine, and C^{14}-sodium formate were the substrates used in these studies. Table 1 depicts the results obtained in this investigation.

These data indicate that carbon atoms number 7A, 3, 3A, 10, and 2 are oxidized to CO_2 by DL-tryptophan-grown cells. No activity was found in the cell suspension with any of the substrates. The high recovery values suggest that these carbon atoms are completely eliminated from the molecule during the dissimilation of tryptophan by this organism.

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TABLE 1. OXIDATION OF ISOTOPICALLY LABELED SUBSTRATES BY DL-TRYPTOPHAN-GROWN CELLS

Substrate	Corresponding Carbon Atom in Tryptophan Molecule	Per Cent C^{14} Recovered in CO_2
7A-C^{14}-DL-tryptophan	7A	87.7
Carboxyl-C^{14}-anthranilic	3	95.3
1-C^{14}-anthranilic	3A	95.8
Carboxyl-C^{14}-DL-alanine	10	84.7
C^{14}-sodium formate	2	99.9

SUMMARY

Experiments using isotopically labeled substrates show that carbons 7A, 3, 3A, 10, and 2 of the tryptophan molecule are oxidized to carbon dioxide by a *Flavobacterium* sp. capable of using either D- or L-tryptophan as a source of carbon and energy for aerobic growth.

LITERATURE CITED

Martin, J. R. and N. N. Durham. 1961. Microbial oxidation of D-tryptophan. Arch. Biochem. Biophys. (In Press).

Stanier, R. Y. and O. Hayaishi. 1951. The bacterial oxidation of tryptophan: A study in comparative biochemistry. Science 114: 326-330.