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PHENAZINE-1,6-DICARBOXYLIC ACID — AN INTERMEDIATE IN PHENAZINE BIOSYNTHESIS?

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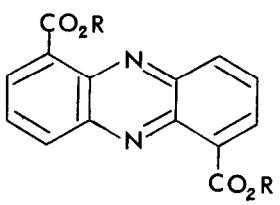
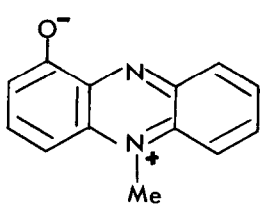
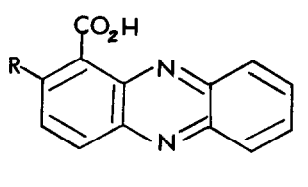
Consideration of structural relationships between the various microbial phenazines, in association with evidence on the manner of shikimic acid incorporation,[1] leads to phenazine-1,6-dicarboxylic acid (1) as a likely common intermediate. Labelled (1) has been found, however, not to be incorporated into pyocyanin (3) in Pseudomonas aeruginosa,[2] phenazine-1-carboxylic acid (4) in P. aureofaciens,[3] or iodinin (6) in Brevibacterium iodinum.[4] Further, we have been unable to trap (1) during a feeding experiment with [^{14}C] shikimic acid in P. aureofaciens. On the other hand, a high (7.4%) and specific incorporation into (4) of dimethyl phenazine-1,6-dicarboxylate (2) has recently been recorded.[3] We have repeated these experiments in P. aureofaciens with (2), labelled with ^{14}C in the carboxy-groups, under various conditions, but have failed to record a total incorporation in excess of 0.2% into the two phenazines, (4) and (5), produced by the organism.[5] We must conclude therefore that dimethyl phenazine-1,6-dicarboxylate (2), like the diacid, cannot act as a significant phenazine precursor in P. aureofaciens at least.



(1) R=H
(2) R=Me



(3)

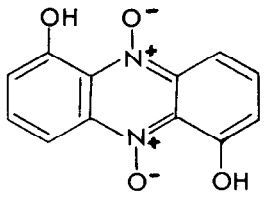


(4) R=H
(5) R=OH

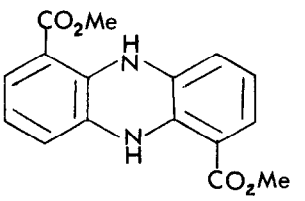
Further consideration of phenazine biosynthesis from shikimic acid leads to hydro-derivatives of (1) as potential intermediates. The dihydro-derivative (7) is

readily accessible by hydrogenation of (2) and sufficiently stable for a biosynthetic experiment. It was examined as a precursor for (4) and (5) in P. aureofaciens, but was found not to be incorporated to a significant extent (0.2%).

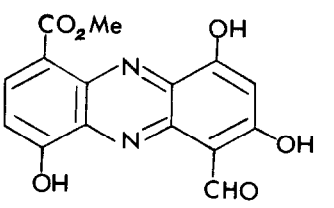
In spite of negative results with (1), (2), and (7) as precursors for phenazines having either one aryl-C_1 substituent [as (4)] or none [as (6)], it was still an attractive possibility that phenazine-1,6-dicarboxylic acid (1) might be a precursor for



(6)



(7)



(8)

metabolites with two aryl-C_1 substituents [as (8)]. Examination of (1) as a precursor for lomofungin (8), in Streptomyces lomodensis, immediately gave positive results. In duplicate experiments satisfyingly high incorporations (6.7 and 9.0%) were observed (assayed on the penta-acetate by recrystallization to constant activity). Phenazine-1,6-dicarboxylic acid (1) is thus clearly a lomofungin (8) precursor, a result of importance to phenazine biosynthesis in general. We are currently investigating whether (1) is a precursor only for phenazines with two aryl-C_1 substituents [as (8)] or whether, simply, it is transported across the cell walls of Streptomyces species but not those of Pseudomonas and Brevibacterium species.

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References

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4. R. B. Herbert, F. G. Holliman, and P. N. Ibberson, J.C.S. Chem. Comm., 355 (1972).
5. Dr. J. M. Turner, of Liverpool University, has kindly informed us that he has obtained similar negative results.