ACCUMULATION OF S-ADENOSYMETHIONINE BY MICROORGANISMS

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ABSTRACT

GAWEL, L. J. (Oregon State University, Corvallis), J. R. TURNER, AND L. W. PARKS. Accumulation of S-adenosymethionine by microorganisms. J. Bacteriol. 83:497–499. 1962.—A series of microorganisms selected to represent a variety of microbial types was cultured under conditions conducive to the formation of S-adenosymethionine. Of the organisms tested, only Saccharomyces cerevisiae accumulated measurable amounts of S-adenosymethionine. Pseudomonas fluorescens and Escherichia coli accumulate 5′-methylthioadenosine under the same conditions.

S-adenosymethionine has been shown to participate in a variety of enzymatic group-transfer reactions, mainly as a donor of methyl groups in transmethylation and as a source of the propylamino group in spermine formation. An excellent review of this subject has recently been published (Shapiro and Schlenk, 1960). It may also function as a sulfur source in cysteine biosynthesis by certain mammalian systems (Stekol, Anderson, and Weiss, 1958). Although S-adenosymethionine (Fig. 1) has been synthesized chemically (Baddiley and Jamieson, 1954) and enzymatically (Cantoni, 1953), the most readily available source has been by direct isolation from yeast previously cultured on methionine-enriched media (Schlenk, Dainko, and Stanford, 1959). Candida utilis may yield up to 20 μmoles of S-adenosymethionine per g, using the enrichment method of Schlenk and DePalma (1957); the ethyl sulfonium derivative, S-adenosylethionine, is also accumulated by this organism (Parks, 1958). The appearance of such large amounts of S-adenosymethionine in these organisms prompted a survey of some other organisms to determine if other cells possess this ability to accumulate S-adenosymethionine.

MATERIALS AND METHODS

Organisms used in this study were obtained from the culture collection maintained in this laboratory. All cells were cultured in a medium consisting of 0.3% beef extract (Difco), 0.5% peptone (Difco), 0.15% DL-methionine, and were supplemented with 1% glucose. Cultures were incubated at 30°C for 72 hr on a rotary shaker. Then the cells were harvested by centrifugation and washed twice with cold distilled water.

The S-adenosymethionine was isolated by a combination of the methods of Schlenk et al. (1959) and Cantoni (1958). The procedure consists essentially of extraction of the packed cells with perchloric acid, precipitation with Reinecke salt, solubilization in methyl ethyl ketone, and removal of the precipitant from an acid solution with diethyl ether. This procedure was designed to recover S-adenosymethionine; however, small amounts of methionine always appeared on the chromatograms. The volume of extraction solvents was adjusted to compensate for differences in cell yields obtained from the growth medium.

Adenine content of the solutions was determined by ultraviolet light absorbance at 260 nm in a Beckman model DU spectrophotometer. Chromatograms were done on Whatman no.1 filter sheets, and the papers were developed in n-butanol-acetic acid-water (60:15:25; v/v). Separated components were revealed by first surveying to observe ultraviolet light quenching by the adenine derivatives; duplicate sheets were then sprayed with either ninhydrin or chloroform reagents.

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RESULTS AND DISCUSSION

Of the ten microorganisms tested, only the yeast accumulated measurable amounts of S-adenosylmethionine (Table 1). The degradation product, 5'-methylthioadenosine (Parks and Schlenk, 1958), was accumulated by Pseudomonas fluorescens and Escherichia coli; an unidentified sulfur-containing derivative was formed by the Arthrobacter organisms. Although large amounts of S-adenosylmethionine were found in the yeast culture, no 5'-methylthioadenosine was detected on the chromatograms. Cell-free preparations of Aerobacter aerogenes, E. coli, and Saccharomyces cerevisiae possess enzymes that rapidly degrade S-adenosylmethionine to 5'-methylthioadenosine and homoserine (Shapiro and Mather, 1958). However, breakdown of the accumulated S-adenosylmethionine by Saccharomyces cells is apparently very limited. Since the S-adenosylmethionine that is stored in yeast is found mainly in the vacuole of the organism (Svihla and Schlenk, 1960), it is undoubtedly protected from degradation.

The high level of S-adenosylmethionine in yeast suggests that the synthetic enzyme is either nonrepressible or that the product, when accumulated in the vacuolar structure, is unavailable to act in inhibition of its own synthesis. In this regard, the S-adenosylmethionine may act as a rather unique cellular storage component containing carbon, nitrogen, and sulfur. Any of these could be readily available from S-adenosylmethionine when growth-limiting conditions exist.

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LITERATURE CITED


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