Effect of Temperature on the Growth of Myxococcus xanthus

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Received for publication 22 December 1976

The cardinal growth characteristics of Myxococcus xanthus were examined from 14 to 40°C, and the examinations indicated that the organism is mesophilic in character. The maximum growth rate (0.3 doublings per h) was between 34 and 36°C and the temperature characteristic (μ) is 17,000 cal/mol (71,162 J/mol).

Under appropriate environmental and nutritional conditions. Myxococcus xanthus (order Myxobacterales) undergoes a series of developmental events in which vegetative cells aggregate and construct a raised mound of cells called a fruiting body (2). Within the fruiting body, many of the vegetative cells convert to resting cells called myxospores. Alternatively, under other appropriate conditions, M. xanthus is capable of exponential growth, and it replicates by binary transverse fission (1). Preliminary results in our laboratory have indicated that differing temperature optima are involved in the complex sequence of events in the developmental life cycle. We report here the temperature range and the cardinal growth characteristics of vegetative growth.

The generation time, or the time required for a doubling of optical density at each temperature, was calculated from the slope of semilogarithmic plots of optical density versus time. The average generation times found at various temperatures are shown in Fig. 1.

Comparison of the vegetative growth range of M. xanthus (Fig. 1) with that of Escherichia coli, as reported by Ingraham (4), shows a similar type of curve and suggests M. xanthus to be mesophilic in character. The absolute minimum for exponential growth was not determined; however, at the lowest temperature examined (i.e., 14°C), the generation time was 25 h, and growth was exponential. The maximum growth rate under the conditions utilized was between 34 and 36°C and was 0.3 doublings per h. At 37°C, the cells grew as filaments, and, at 39°C and above, cellular morphology was aberrant with considerable spheroplasting. The absolute temperature maximum is about 40°C. This temperature range of growth is narrower than that of E. coli, which is capable of growth within the approximate range of 8 to $47^{\circ}C$ (5).

Figure 2 is an Arrhenius plot where the natural log of the growth rate is plotted against the reciprocal of the absolute temperature. The



FIG. 1. Effect of temperature on the generation time of M. xanthus. Cells were grown in liquid media consisting of 2% Casitone (Difco), 10 mM potassium phosphate buffer (pH 7.6), and 8 mM magnesium sulfate. M. xanthus strain MD-1 (previously referred to as strain FB) was used throughout this study. Cells were grown in a large culture tube (200 by 40 mm) submerged in a Lauda K/2R circulating water bath $(\pm 0.2^{\circ}C)$. Air was water saturated, heated, or chilled to the culture temperature and passed through two Pasteur pipettes submerged in the culture. Growth under these conditions was identical with growth under optimal conditions in a rotating shake culture. Mass doubling was followed at each temperature by means of a Klett-Summerson Colorimeter with a no. 54 filter, and measurements were taken in the range of 50 to 200 Klett units. Throughout this range, the culture is in exponential phase, and there is a direct relationship between cell mass and the amount of light scattering detected by the Klett. Klett data were plotted against time, and the best-fit line through these points was calculated. The mass doubling time was then calculated directly from the slope of this line.



FIG. 2. Arrhenius plot relating growth rate and temperature in M. xanthus. Growth rate is in terms of generations per hour.

temperature characteristic (μ) obtained for *M*. *xanthus*, calculated from Fig. 2, was 17,000 cal/mol (71,162 J/mol).

It has been suggested that the value of μ is a definitive characteristic of a given species, with which it could be classified as a mesophile or a psychrophile (4). However, most recent evidence (3, 6, 7) seems to indicate that the μ obtained for an organism is not necessarily related to the temperature range in which the

organism is capable of growth. The significance of μ as a descriptive parameter is unclear.

With the data presented here, one can readily determine that the useful range for temperature-sensitive mutant studies is 26 to 35° C. It would also be of interest, from an ecological point of view, to examine the effect of temperature on the various aspects of the developmental life cycle of *M. xanthus*.

This work was supported by Public Health Service grant GM-19957 from the National Institute of General Medical Sciences (to M.D.).

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