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Abstract

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Mannitol non-utilizing mutants were isolated following UV irradiation of wild type 74-OR23-1A (74A) conidia and filtration concentration in the minimal medium of Westergaard and Mitchell (W-M) in which mannitol was the sole carbon source. A cross of one of these mutants to wild type 74-OR8-la (74a) yielded a progeny, M14-62, that was colonial and had a gummy appearance. When grown at 25° C in constant light, M14-62 produced concentric rings in plates and bands in race tubes. Examination with phase-contrast microscopy showed that the pattern of growth was due to periodic, increased hyphal branching. The pattern of banding ceased to be seen in cultures incubated in constant dark for an extended period of time.

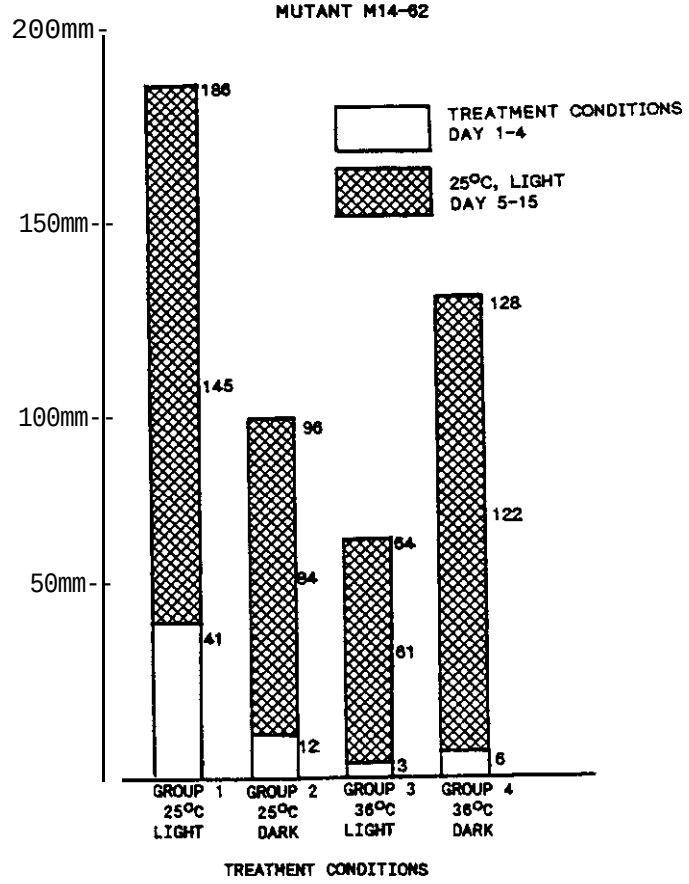
The morphology of M14-62 and the fact that it was derived from a mutant unable to grow on mannitol which is a well known osmoticum, suggested the possibility that it might be an osmotic-sensitive mutant. Growth tests indicated that M14-62 was indeed osmotic-sensitive since it did not grow on medium containing 6% NaCl, one of the criteria for identifying such mutants (Mishra 1977 Genet. Res. Camb. 29:9-19). In crosses of Ml4-62 to 74a, osmotic sensitivity segregated 1:1 (60 mutant and 52 wild type progeny). Morphology also segregated 1:1 (48 mutant and 64 wild type progeny). Two progeny had mutant morphology but grew on salt medium; 14 progeny had wild type morphology but did not grow on salt medium. It is not known whether these 16 progeny are truly recombinant or if modifier genes might be present.

The mutant M14-62 was sensitive to elevated temperatures. In petri plates containing W-M medium, the average diameter of eight day cultures at 31° C was 19 mm compared to 24 mm at 25° C, a reduction of 21%. When the cultures were incubated at 37° C, the diameter was 12 mm, a reduction of 50%.

To test the effects of light-dark and temperature on M14-62, race tubes containing W-M medium were inoculated with M14-62 conidia from cultures which had been kept in constant light for more than six months. The tubes were divided into four groups. Light-treatment tubes were uncovered. Dark-treatment tubes were wrapped in aluminum foil, care being taken not to cover the cotton plugs to avoid altering gas exchange. Uncovered tubes (Group 1) and covered tubes (Group 2) were placed at 25°C. Similar tubes, uncovered (Group 3 and covered (Group 4),were placed at 36° C. In each experimental condition, wild type 74A served as a control. After four days, the foil was removed from the dark-treatment tubes and the growth of each group was measured. All tubes were then placed at 25°C in the light. After 11 days in constant light at 25° C. the growth in each tube was again measured.

All wild type 74A controls had reached the end of the race tubes by day four. Results of the experimental M14-62 cultures (Groups 1,2,4-average of 4 trials; Group 3 - average of 3 trials) suggest an interaction of light-dark and temperature on the growth of M14-62 (Fig. 1). At 25° C, growth in the dark was retarded by 70% compared to growth in light. Absence of light for the first four days of culture also retarded subsequent growth in light hy more than 40% (Group 2 vs. Group 1). Although during the first four days at 36° C, Group 4 (dark) grew more than Group 3 (light), (6mm vs. 3mm), the very restricted growth at this temperature makes it difficult to attribute the small difference to the dark growth conditions. However, exposure to continuous light at 36° C for the first four days of

Figure 1. THE EFFECT OF TEMPERATURE AND LIGHT ON GROWTH OF THE OSMOTIC SENSITIVE MUTANT M14-62



culture (Group 3) seemed to have a lasting effect for when these cultures were placed at 25° C in continous light, Group 3 had reduced growth compared to all other treatment groups. In contrast, those cultures which had been in the dark at 36° C for the first four days (Group 4) seemed to grow almost as well as Group 1 after being moved to light at 25° C.

It appears that in the mutant M14-62 light-dark and temperature interact to exert an effect on growth which continues even after the mutant is transferred to ambient temperature and continuous light. - - - Dept. of Biology, Georgia Southern College, Statesboro, GA 30460