A Study of Variants of Nocardia corallina¹

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Variation is one of many notable factors contributing to the difficulty encountered when studying the *Actinomycetes*. This preliminary study was undertaken to determine the differences that might exist among several available variants of *Nocardia corallina* (ATCC 4273). These organisms differed from the parent strain in color and/or in colonial morphology. This study compares the characteristics of the variants to those of the parent strain on the basis of morphology, physiology, and rate of the growth cycle, hyphal length and fragmentation time.

The organisms were selected from a large number of variants isolated during cytogenetic studies using the 4273 strain. The variants were cultured on nutrient agar + 1% fructose and incubated at 29 C. The growth cycles were followed by inoculating the organisms onto agar slants and, at regular intervals, transferring small amounts of the surface growth to alides which were then stained to demonstrate nuclei, according to the method of Chance (1952).

All organisms were also tested for the possession of acid-fastness, diastatic and proteolytic activity, and the ability to reduce nitrates and to produce acid and/or gas from standard carbohydrate media. The variants showed no significant differences from the parent strain in the physiological studies, acid-fastness and nitrate reduction.

The color of the typical N. corallina is pink, but the colors of the variants ranged from a distinct red, through pink to white. The typical strain normally grows as predominately large, rough, wrinkled, folding colonies with rhisold-type margins.

When a culture of N. corallina 4273 is streaked on agar plates, a certain percentage of the colonies will grow as smooth, dull-surfaced variants. Webb (1956) calculated this number to be between 0.01 and 0.3% of the total number of colonies present. The organisms used in this study and labeled as smooth variants yielded from 90 to 100% smooth colonies.

A comparison of growth cycles of the rough and smooth cultures showed a consistent difference. The fragmentation time of the 4273 strain was 14 to 16 hours and the hyphae reached an average length of 10 to 15 μ . In the smooth variants, the hyphae were much shorter, around 7 to 9 μ , and the fragmentation time was 10 to 12 hours. Colonies of some of the variants were larger and possessed an even more wrinkled, folded surface than the parent strain. The hyphae in these organisms were longer, some 20 to 25 μ , and also the fragmentation time was longer, up to 25 hours in duration.

This preliminary work indicates that the only significant difference,

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other than color in N. corallina ATCC 4273 and the variants which were studied, lies in the fragmentation time and hyphal length in the growth cycle, which can be correlated with the roughness and smoothness of the growth on solid media. McClung (1954), in his study of utilization of carbon compounds by Nocardia species, concluded that this particular characteristic is probably stable and distinct enough to be used as one of the major methods for separating the species of this genus. With the species studied, he found no complete duplication of utilization results. The 7th Edition of Bergey's Manual (1957) uses the color of growth as the initial factor for separating several Nocardia species within the same subgrouping, e.g., N. corallina, N. rubropertinota, N. rubra, and N. coeliaca.

Because of the normal variability that occurs within a single species, e.g., color and rough and smooth variation, it is confirmed that identification of *Nocardia* species should perhaps be based primarily on physiological reactions and, to a lesser extent, on cultural morphology. However, variation in this group must receive more study in order to fully elucidate the group and must be considered in any taxonomic scheme.

LITERATURE CITED

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