FACTORS TO BE CONSIDERED IN THE
CLASSIFICATION OF ACTINOMYCETES*

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The important factors to be considered in regard to the classification of actinomycetes pertain primarily to the recognition of genera and species. Group characteristics are only of secondary importance.

Recognition of genera

All the accumulated evidence points to the recognition, at the present time, of 10 genera in three families of the class Actinomycetales, namely, the Actinomycetaceae, the Streptomycetaceae, and the Actinoplanaceae. These can be classified as follows:

A. Spores formed, but not in sporangia.

I. Substrate mycelium fragmenting into bacillary or coccoid elements.

Family I. Actinomycetaceae Buchanan

1. Anaerobic or microaerophilic, parasitic, not acid-fast. 1. Actinomyces Harz.
2. Aerobic, some parasitic, partially acid-fast or nonacid-fast. 2. Nocardia Trevisan.

II. Substrate mycelium nonseptate, not fragmenting into bacillary or coccoid elements.

Family II. Streptomycetaceae Waksman and Henrici

1. Aerial mycelium not produced.
 - a) Spores formed singly on short sporophores.
 - a₁. Mesophilic forms.
 3. Micromonospora Ørskov
 - b₁. Thermophilic forms.
 4. Thermomonospora Henssen

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2. Aerial mycelium produced.

- a) Spores formed in chains; mesophilic and thermophilic forms.

5. Streptomyces Waksman and Henrici

- b) Spores formed singly; thermophilic forms.

6. Thermoactinomyces Tsiklinsky

- c) Spores formed in pairs.

a₁. Mesophilic forms.

7. Waksmania Lechevalier and Lechevalier (Microbispora Nonomura and Ohara)

b₁. Thermophilic forms.

8. Thermopolyspora Henssen

B. Spores formed in sporangia.

Family III. Actinoplanaceae Couch

- I. Aerial mycelium usually not formed, coiled conidiophores lacking, sporangiospores motile.

9. Actinoplanes Couch

- II. Aerial mycelium abundant; coiled conidiophores as well as sporangia formed in some species, sporangiospores nonmotile.

10. Streptosporangium Couch

Recognition of species

The problems involved in species differentiation of actinomycetes, especially within the genus Streptomyces, must take into consideration primarily morphological and cultural criteria. Some of the biochemical characteristics offer supplementary information. These properties may be briefly summarized as follows.

1.) Morphology includes the structure of the colony and of the substrate mycelium; the nature of the aerial or surface mycelium and manner of sporulation; the size, shape, and surface structure of spores.

2.) Cultural properties include growth characteristics on certain organic and synthetic media. Solid media are primarily used. The common liquid media do not offer much supplementary information. Pigmentation of the aerial and substrate or vegetative mycelium, formation of soluble pigments, proteolytic activities, notably gelatin liquefaction and milk coagulation and proteolysis, as well as growth

BACTERIOLOGICAL NOMENCLATURE AND TAXONOMY

on potato and on certain other selected media, are most important.

3.) The biochemical activities include, in addition to the above, sucrose inversion, diastatic and reducing properties, utilization of different carbon sources, and formation of antibiotics.

4.) Sensitivity to specific phages, and, to some extent, serological reactions, offer additional specific characteristics.

5.) Standard media must be adopted for purposes of characterization. These comprise both well-defined solid synthetic media and carefully selected organic solid and liquid media. Conditions of growth for the purpose of species characterization should be constant.

6.) Certain important characteristics should also be recognized for establishing strains or varieties of organisms within a single species. These are usually based upon morphological and cultural variations, upon the ability to produce economically important substances, such as vitamins, enzymes, and antibiotics, and upon other properties, such as cross resistance, phage and antibiotic sensitivity.

7.) Type cultures should be properly preserved, deposited in recognized culture collections, and made available to all investigators throughout the world.

8.) No scientific journals should accept contributions in which a new species is listed without a proper description of the organism in a recognized international language or a reference to a previously published description, and without its prior deposition in a recognized culture collection.

Recognition of groups

It has been recognized, since the early days of the study of actinomycetes, that some of the forms, now found to belong largely to the genus Streptomyces, represent groups of organisms rather than single species. Sanfelice, as far back as 1904, suggested the division of the genus, designated as Streptothrix, into three groups: S. alba, S. flava, and S. violacea; it is quite evident from these designations that pigmentation was used as the basis for group separation. Waksman (1919) also recognized the need for separating the aerial mycelium-producing actinomycetes into species-groups, on the basis of their morphological and cultural properties. He demonstrated three morphological types,

those forming straight sporulating hyphae, spiral-shaped hyphae, and verticil-forming organisms. He stated "often several strains of the same 'species-group', when compared on the same culture medium might be taken culturally and often morphologically as distinctly different species."

More recently, Baldacci et al. (1954) suggested a division of the organisms belonging to the genus Streptomyces into "sections" and "series," largely on the basis of color of the substrate and of the aerial mycelium, respectively. Each "series" was to be divided into species on the basis of enzymatic activity, antibiotic properties, and soluble pigments. Twenty-one series were thus recognized. This grouping was later variously modified by Baldacci (1956).

Gause et al. (1957) presented a rather elaborate system of classification of the genus Streptomyces using a set of rather peculiar assumptions. On the one hand, the generic name Streptomyces was not recognized officially; on the other hand, however, reference to all other species, usually included under the genera Actinomyces, Micromonospora, and others, was excluded, thus automatically recognizing the genus Streptomyces by an omission which consisted in carefully avoiding this name.

The separation of the genera into groups is based, first of all, on the pigmentation of the substrate growth and of the aerial mycelium; some of the morphological properties of the organisms, notably manner of sporulation and nature of spores, are also taken advantage of in the separation of the genus Streptomyces into groups. It must, of course, be kept in mind that any attempt to separate the genus Streptomyces into groups will accomplish no more than the raising of some of the important species to subgeneric status.

Variability

Any system of classification of such a variable group of microorganisms as the streptomycetes must recognize the continuity between different species. Allowance must also be made for transitional forms which would tend to bridge the gaps between the individual species. The need for a natural system of classification is as great for the Actinomycetales as for any other group of bacteria. One must consider, first of all, the morphological properties, especially the formation of aerial mycelium and the type of sporulation. Then come the cultural properties, notably

BACTERIOLOGICAL NOMENCLATURE
AND TAXONOMY

pigment formation and growth upon different media. Consideration must also be given to the ecological properties, notably anaerobism vs. aerobism, pathogenicity vs. saprophytism and thermophily vs. mesophily. There are, further, the physiological activities, including carbon and nitrogen utilization. Finally, there are the chemical properties (composition of cells) and the biochemical activities, such as the property of formation of antibiotics, enzymes, and vitamins. All of these can serve as supplementary characteristics for describing species and varieties of actinomycetes.

Recommendations

It is proposed to utilize a combination of ecological, morphological, and certain physiological properties of the organisms for the characterization of genera of actinomycetes. Some of the morphological, cultural, and physiological properties may be used in establishing groups within the genera. Cultural and biochemical properties are utilized for characterizing species. Finally, the recognition of varieties must be left to the individual investigators, who may consider such problems as variability and specific biochemical activities, such as sugar utilization and antibiotic production.

Formation of antibiotics should certainly be considered among the properties for species characterization. Carbon sources as a specific property is open to question; it may be left to varietal characteristics.

ADDENDUM

Proper use of the vernacular words "actinomyces" and "actinomycetes", "streptomyces" and "streptomycetes"

Ever since Harz gave the first valid description of an actinomycete in 1877, there has been considerable confusion as to the nomenclature of this group of organisms. There are certain reasons for this confusion: 1. The close similarity between the scientific name Actinomyces and the vernacular names "actinomyces" and "actinomycete" has tended to confuse the workers in this field. 2. Especially since 1943, when Waksman and Henrici proposed the new genus

Streptomyces, which included many, but by no means all, of the species previously assigned to the genus Actinomyces, the situation has tended to be further confounded.

Recent literature on the actinomycetes is replete with many examples of the use of the words "actinomyces" and "Actinomyces" where it is virtually impossible to determine whether the author is referring to all of the actinomycetes, to the genus Streptomyces, or to the genus Actinomyces. In fact, the writer readily admits to such confusing terminology in some of his own publications. To avoid at least some of this difficulty, the writer makes the following proposals:

1. The name "actinomyces" may be used as the vernacular expression only for members of the genus Actinomyces; it should no longer be considered as synonymous with the singular "actinomycete" or the plural term "actinomycetes." Likewise "streptomyces" is the acceptable vernacular name when used to refer solely to members of the genus Streptomyces.
2. The vernacular terms "actinomycete" and "actinomycetes" can properly be used as inclusive terms for any or all of the organisms now included in the Actinomycetales, exclusive of the mycobacteria and corynebacteria, without referring to either families or genera. The words "streptomycete" and "streptomycetes" should be limited to members of the family Streptomycetaceae.