PROPOSED PHYLOGENY IN THE
FAMILY BACILLACEAE

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The family Bacillaceae enjoys the distinction among true bacteria of having a readily identifiable, taxonomically acceptable, characteristic in the endospores which serve to differentiate it from other families of the Eubacteriales. The large number of distinct and well characterized species allow comparison of species characteristics within the family. It provides a logical starting point for a study of a segment of bacterial phylogeny and gene flow that may through application of appropriate experimental design ultimately establish criteria for better understanding of relationships within the entire order. The demonstration of nuclear interchange in several species of bacteria as well as indirect evidence of its occurrence in the genus Bacillus (Delamater, 1953; Johnson, 1960) support this theoretical approach.

In reviewing the taxonomy of this family (Breed et al., 1957), it is apparent that nitrogen fixation, cellulose digestion and exotoxin production are not combined in any known species of the genus Clostridium. While most species in these three groups are anaerobic and non-pigmented, each possesses at least one species that is pigmented, and two of the three have at least one that is a facultative aerobe, suggesting a relationship in these two directions.

Since strict aerobes and strict anaerobes are logically and probably ecologically incapable of direct nuclear transfer, facultative members of both Bacillus and Clostridium were checked for intermediary characteristics.

Sub-criteria used in this analysis were: acid and gas from carbohydrates, acid only, no acid or gas from carbohydrates (without reference to specific sugars), gelatin liquefaction, acetyl-methyl-carbinol production, swollen or non-swollen sporangia, Gram stain reaction and optimum temperature.

Figure 1 shows the postulated relationship between species of the family, using key species for intermediates in the larger groups.

BACTERIOLOGICAL NOMENCLATURE AND TAXONOMY

If strict anaerobes are considered primitive, the pigmented Clostridia seem to include the necessary characteristics for giving rise to three distinct groups of species, i. e. cellulose digesters, nitrogen fixers and exotoxin producers. The major characteristics of this pigmented group are: swollen sporangia, acid and gas from sugars, gelatin not liquified, temperature optima, Gram reaction negative to variable, and mesophilic.

The cellulose digesters are primarily: Gram-negative with swollen sporangia, gelatin negative and attack carbohydrates weakly or not at all.

The nitrogen fixers are primarily: Gram-positive with swollen sporangia, attack sugars with production of acid and gas, gelatin negative and mesophilic.

The exotoxin producers are primarily: Gram-positive with swollen sporangia, produce acid and gas from sugars, liquefy gelatin and are mesophilic.

Six distinct lines of development can be shown from these three major groups.

1. From the thermophilic cellulose-digesters through facultative aerobic thermophiles to the facultative thermophile Bacillus coagulans.
2. From the cellulose-digesters with non-swollen sporangia to B. coagulans.
3. From the facultatively aerobic nitrogen-fixers through Clostridium lacunarum to B. cereus.
4. From the exotoxin-producers with non-swollen sporangia through the antibiotic producers B. licheniformis and B. subtilis to B. cereus.
5. From the facultatively aerobic exotoxin-producers attacking sugars with acid and gas formation through B. polymyxa to B. cereus and B. circulans.
6. From the facultatively aerobic exotoxin-producers not attacking sugars through B. pasteurii to both B. cereus and B. sphaericus.

The work of Smith et al. (1946) has done much to clarify the interrelationships among the species of Bacillus. Their work indicated that B. coagulans, B. cereus and B. circulans are species with variable characteristics as might well be expected of species playing the intermediate role here assigned them.

The strict aerobes (genus Bacillus) in the family have in common inability to produce acetyl-methyl-carbinol, liquefy

gelatin (except B. lentus) and fail to attack sugars (except B. megaterium - acid, no gas). Taxonomic intermediates between B. cereus and B. megaterium are well known as well as the close relationship of B. pasteurii and B. sphaericus. B. circulans is viewed as giving rise to both B. lentus and B. brevis.

This analysis suggests that Gram-negativeness, pigment production, spores in swollen sporangia, non-liquefaction of gelatin, anaerobiosis, acetyl-methyl-carbinol production and acid and gas from sugar may be primitive characteristics of this family. Clostridium venturelli has these characteristics (VP reaction not reported). The converse of these reactions would represent the most advanced species (B. firmis).

That cellulose digestion, nitrogen fixation and exotoxin production occupy similar or identical gene loci is suggested. Except for the relationship of strict aerobiosis to lack of acetyl-methyl-carbinol production, all characteristics used here appear to operate independently.

The author realizes that modern phylogeny envisions a reticulate rather than mono-linear form of development. However, the present status of bacterial phylogeny suggests beginning with the latter in hopes that the former may be evolved to provide the field with a more definable concept of bacterial species.

Other criteria reported in the literature (Bradley and Franklin, 1958; Salton, 1956; Judith and Weinberg, 1959) have been used and are in general agreement with the scheme here given. However, the species covered by these authors have been too few to employ them as general criteria for this study.

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