# Taxonomy in Phytophthora

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For some time it has been obvious in all groups of plants that delimitation of taxa on morphological grounds alone is not enough, especially with controversial groups and those that have few morphological characters. Although in the Code of Botanical Nomenclature it is not specified that taxonomy and nomenclature should be based on morphology, it has been tacitly assumed in the past that this was so. At present we must accept morphology as a basis in *Phytophthora* until better methods have been found, but it is now essential to supplement it with other criteria.

The taxonomy of *Phytophthora* needs clarification and the newer investigational methods should be useful. Taxonomy is based on the type method (bound by the Code of Botanical Nomenclature), and at present this method should be followed, as the specific epithet is indissolubly tied to the type. A type, be it the original specimen, culture, or description with figures, or all three, must be the basis of classification. Unfortunately no original type specimens exist for many species. I have hesitated to designate new types, permissible under the Code, where none are known, because of the possible instability of cultures and the unsatisfactory nature of dried specimens for *Phytophthora*. If new types are designated they should be obtained if possible from the type locality and from the type host or habitat, and should conform as nearly as possible to the original descriptions and figures. An essential starting point, always, is the original; until this has been studied one's concept of a taxon may be incorrect in some detail or details. No name on a test tube should ever be accepted until the contents have been compared with the type isolate and/or description. If an isolate differs in any way from this norm the differences should be pointed out so that future investigators will know exactly what was used; if the differences are great the epithet should not be applied without some qualification.

Of morphological criteria that have been used in the past, one which has been discredited to a certain extent is cultural form which depends on the frequency, angle, and extent of branching of the hyphae. This is very little understood, and no work so far as I am aware has been done in *Phytophthora* on the factors underlying this. It appears to be a notoriously variable character and possibly under cytoplasmic control (4), but even so certain species do have a cultural pattern which persists under a variety of cultural conditions (e.g., the rose pattern of *P. syringae,* the chrysanthemum pattern of *P. citricola,* and the very tough, fluffy uniform mat of *P. cinnamomi*). In recent genetical studies, cultural patterns and hyphal branching were used as markers (9), and they appeared to be stable into the next generation. Therefore, cultural characters should not be dismissed without further work.

One of the newer taxonomic aids in the morphological field is ultrastructure. Electron micrographs give more detailed pictures of structures that are doubtfully distinct under the ordinary microscope. Only about four *Phytophthora* species appear to have been investigated, and only parts of the life cycle. Chapman & Vujičič showed (5) that in *P. erythroseptica* the sporangium is entirely covered with an outer wall layer continuous over the apex; that when the sporangium is chilled to initiate zoospore formation, a vesicular layer develops inside the outer wall layer as zoospore development proceeds (it is thicker under the apex); apical thickening is narrow and unlaminated; during preparation for discharge another wall layer develops inside the vesicular layer; an evanescent vesicle is formed from the vesicular layer; no flagella appear before cleavage; and in the young sporangium there is a central glycogen vacuole.

In contrast, in *P. infestans* (14) the outer layer is not continuous over the apex; the apical thickening fills the gap and makes the apex papillate; there is no vesicular layer all round inside the wall but it develops only beneath the apical thickening; the latter is deep and laminated; there is no evanescent vesicle (the layer from which it arises in *P. erythroseptica* is not present); flagella are present long before cleavage; and there is no vacuole in the young sporangia, though a nonglycogen one develops in sporangia too old to form zoospores.

Hohl & Hamamoto (13) found sporangial structure in *P. parasitica,* another papillate type, to be essentially the same as King et al. (14) found in *P. infestans* except that the outer wall layer appeared to be continuous over the apex and that an evanescent vesicle developed from the outer layer of cytoplasm.

These details of structure support the view that

forms with papillate and nonpapillate sporangia are distinct. Recently, Galindo & Zentmyer (9) crossed two nonpapillate isolates and obtained papillate forms from germinating oospores. One would like to know whether the fine structure of parents and progeny follows the pattern of *P. erythroseptica* and *P. infestans*. The authors do not record in their paper whether they were astonished at this result, but they cannot have been more astonished than Punnett (17), who crossed two races of white sweetpea (so eliminating, he thought, any embarrassment with color) when examining two types of standard, upright and hooded. In the $F_1$ all the progeny were colored. The explanation was that two or more factors were needed for color; they were separated in the parents and came together in the progeny. The same may well be true for nonpapillate and papillate characters.

Further electron microscope work on the base of the sporangium should show differences between deciduous and nondeciduous sporangia, and between proliferating and nonproliferating. Investigation of oogonial and oospore walls also may reveal differences between species.

Sporangium shape has been severely questioned as a factor of taxonomic value. It is, however, an expression of two fundamental opposing forces, one arresting growth in the apical direction and the other causing a lateral expansion which is different at different points along the axis.

Cytological investigations have shown some chromosome numbers: approx. 10 in *P. cactorum* (19), 9 in *P. erythroseptica* (18), 8 in *P. drechsleri* (9), and 8-10 in *P. infestans* (15), none sufficiently different to aid taxonomy.

Morphological features that have received little investigation are: paragyny and amphigyny and what governs their appearance; monocliny and dicliny, sporangiophore branching, and chlamydospores. There is a type of amphigyny in strains of *P. nicotianae* in which the oogonial hypha penetrates the side of the antheridium. Consequently, the latter appears to lie at the side of the oogonial stalk (apparently paragynous), whereas in fact the oogonial stalk is just inside. Sometimes in microscopic mounts it is not possible to determine the nature of an antheridium. Therefore, in counts, in addition to the absolutely certain ones, the percentage of doubtfuls should also be recorded.

New aids outside the morphological field are chemical investigations; viz. (i) serology, (ii) electrophoresis of proteins, and (iii) microchemical analyses. Both serology and electrophoresis have demonstrated major protein differences between morphologically distinct species. Burrell et al. (3) obtained such serological distinctions between *P. cactorum, P. citricola, P. cinnamomi,* and *P. erythroseptica*; Merz et al. (16) between *P. cinnamomi* and the *P. palmivora* group. Within the latter group, however, their techniques did not distinguish between isolates which were fairly close morphologically but with oogonia of different sizes. However, other isolates apparently similar and with similar oogonia were serologically different. What is serology telling us?

Preliminary electrophoresis tests by Clare & Zentmyer (6) showed differing enzymic patterns between *P. cinnamomi, P. palmivora,* and *P. citrophthora* (11). Gill & Powell (10) distinguished *P. cactorum, P. fragariae,* and *P. sojae* by this means. This chemical work supports some longstanding taxa.

Microchemical analyses have shown only slight differences between species so far (1), but these differences are worth pursuing with improved or different techniques. Tests of oogonial walls, some of which are thin and some thick, some colorless and some becoming brown, might be a profitable line of investigation.

The use of pathogenicity tests as an aid to taxonomy seems to have faded. Tucker (23) used them to a considerable extent as an additional aid to support his delineation of species. Where specific distinctions become clearer, such pathological differences might be retested.

What now is the position of speciation? The status of some specific taxa is still in question. I agree with Smith (21), who believes in a narrow species concept for the higher fungi, and I would apply it to this genus. In the state of our knowledge at present and in view of the still preliminary nature of the newer approaches reviewed above, it would be best to maintain a halt and refrain from mergings and splittings. The only recent comprehensive morphological appraisal (20) was concerned mainly with the sexual organs. It did not follow closely the type method; existing type cultures, authentic cultures, or in the absence of these, isolates most nearly conforming to type were not clearly designated or used as starting points and bases for comparisons to segregate those isolates which deviated in some way. The work on the whole supported well-established species, but left the notoriously difficult *palmivora* and *nicotianae* groups, to which must be added now the *megasperma* complex, unresolved.

There are two major problems here: variation and the probability that hybrids exist. Variation in *Phytophthora* we must live with. Erwin et al. (8) reviewed the subject comprehensively, but found comparatively little about morphological and cultural variation. In a study of variation in *P. cactorum* by Stamps (22), all the variants fell clearly within the species. Caten & Jinks (4) did not say that their very variable subcultures were other than *P. infestans*. But this may not be true for other species. The fact that variation is common means that the conditions for taxonomic studies must be clearly defined and rigidly controlled. Hendrix (12) and Brasier (2) found that sporangia of *P. palmivora* grown in the light were typical; not surprising since this species sporulates on aerial parts of plants, while those grown in the dark were not. This may explain some of the difficulties in characterizing this species over the years.

Genetical studies of pairings between closely related taxa that have been considered by some workers to be conspecific and by others to be different species may show some to be hybrids. Progeny morphologically distinct from the parents should be compared with similar-named species and back-crossed to them to find which are masquerading as species. Then the correct

hybrid terminology can be applied, as it was in *Allomyces* (7). No good will be served by sinking possible hybrids under a species until they are properly labeled.

Recently some unpublished pages of one of Darwin's notebooks were found. In them appeared this statement: "My definition of species has nothing to do with hybridity" but "is simply an instinctive impulse to keep separate". Unfortunately, it is not possible to explain instinctive impulses to other people, but nevertheless they may be good "hunches". They appear to work in *Phytophthora* sometimes and should not be ignored but tested in every possible way.

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