Phenotypic heterogeneity of *Pseudomonas syringae* van Hall

L. Gardan ⁽¹⁾, S. Cottin ⁽¹⁾, C. Bollet ⁽²⁾ and G. Hunault ⁽³⁾

⁽¹⁾ Station de Pathologie végétale et Phytobactériologie, INRA,
49070 Beaucozéz (France),

⁽²⁾ Hôpital Salvator, 249 Bd Ste Marguerite, 13009 Marseille (France), and

⁽³⁾ Faculté des Sciences, Bd Lavoisier, 49045 Angers Cedex (France)

SUMMARY

The study of phenotypic properties of 108 strains of *Pseudomonas syringae* pv. *syringae* van Hall isolated from Cherry laurel (50 strains) and various host plants (58 strains) and 53 strains of other pathovars of *P. syringae* and fluorescent *Pseudomonas* showed that the majority of the strains (91/108) were clustered in one phenon (phenon 14) containing strains commonly considered as *P.s.* pv. *syringae*. The present type strain of *P.s.* pv. *syringae* was distantly related to phenon 14. Other pathovars of *P. syringae* constituted 13 discrete phenons.

Key-words: *Pseudomonas syringae*; Phenotyping, Pathovars, Numerical taxonomy.

INTRODUCTION

It has been demonstrated that Plant pathogenic oxidase-negative fluorescent *Pseudomonas* giving hypersensitive reaction on tobacco leaves were shown to be distinct from other fluorescent *Pseudomonas* giving no hypersensitive reaction on tobacco leaves, and which are saprophytic or pathogenic for humans and animals (Stanier *et al.*, 1966; Misaghi and Grogan, 1969).

Fluorescent phytopathogenic *Pseudomonas* are responsible for many plant diseases and cause heavy damage and losses; their economic impact is of major importance. Many pathological aspects have been studied in detail, but studies on identification (Lelliott *et al.*, 1966; Hil-

debrand and Schroth, 1972) and taxonomy (Mishaghi and Grogan, 1969; Sands *et al.*, 1970; Pecknold and Grogan, 1973) are limited.

For some time, their identification was based on the schemes of Lelliott *et al.* (1966) and Hildebrand and Schroth (1972), in which 17 species were recognized.

Before 1978, a new strain isolated from a host plant and pathogenic toward this plant was considered as a species. In 1978, Young *et al.* proposed the concept of pathovar for designating strains of a given species which associated with a disease on a distinct host. Bacteria included in group I of Lelliott *et al.* (1966) were considered to constitute one species, *P. syringae*; the type strain of this species pathogenic for the lilac

belongs to the pathovar *syringae*. The list of pathovars of *P. syringae* was published in 1980 (Dye *et al.*, 1980) and completed by Bradbury (1986). The number of pathovars in *P. syringae* is about 52. It is presently impossible to differentiate these pathovars on the basis of biochemical characteristics.

It has not been demonstrated by objective methods whether all pathovars of *P. syringae* constitute a single species. The purpose of the present work was to compare *P.s. pv. syringae* to other pathovars of *P. syringae* by using a large number of phenotypic tests. The outcome shows the phenotypic heterogeneity in *P. syringae*.

MATERIAL AND METHODS

Strains

Three different sets of strains were used:

— 50 strains of *P.s. pv. syringae* isolated from cherry laurel in France between 1976 and 1986 (leaf spots, necrosis and leaf surface), belonging to 5 cultivars ("Caucasica", "Otto Luyken", "Rotundifolia", "Reynvaanii" and "Herbergii") were selected for their geographic, varietal and temporal origins, from a collection of 241 strains;

— 58 strains of *P.s. pv. syringae* isolated in France and in other countries, between 1950 and 1986, from various hosts distributed in 31 botanical genera (table I);

Table I. Origins of strains of *P.s. pv. syringae* isolated from various host plants.

Host	CFBP no.	Origin and year of isolation
<i>Allium cepa</i>	1787	Japan, date unknown
<i>Callistemon viminalis</i>	1776	New Zealand, 1972
<i>Citrus clementina</i>	1607	France, 1975
<i>Citrus limon</i>	1703	Italy, 1968
<i>Citrus sinensis</i>	1779	Greece, 1962
<i>Citrus</i> sp.	1772	USA, 1975
<i>Cotoneaster</i> sp.	1773	France, 1975
<i>Crataegus</i> sp.	1689	UK, 1961
<i>Cucurbita maxima</i>	1788	New Zealand, 1973
<i>Cucurbita</i> sp.	1780	USA, 1967
<i>Delphinium</i> sp.	N12.2 (*)	France, 1980
<i>Euphorbia pulcherrima</i>	1777	New Zealand, 1972
<i>Evonymus japonicus</i>	1542	France, 1974
<i>Forsythia</i> sp.	2576	France, 1979
<i>Forsythia</i> sp.	M10.5 (*)	France, 1979
<i>Hibiscus esculentus</i>	1760	Kenya, 1965
<i>Hibiscus</i> sp.	1758	Australia, 1960
<i>Juglans regia</i>	700	France, 1965
<i>Lycopersicum esculentum</i>	1789	Australia, 1970
<i>Magnolia</i> sp.	1678	UK, 1956
<i>Magnolia</i> sp.	P65.4 (*)	France, 1981
<i>Malus domestica</i>	2559	France, 1980
<i>Malus sylvestris</i>	602	France, 1964
<i>Olea europea</i>	S43.3 (*)	France, 1983
<i>Oryza sativa</i>	1655	Hungary, 1956
<i>Persea americana</i>	1701	New Zealand, 1955
<i>Phaseolus vulgaris</i>	1514	USA, 1966
<i>Phaseolus vulgaris</i>	1690	USA, 1920
<i>Pisum sativum</i>	1765	New Zealand, 1970
<i>Populus canadensis</i> var. <i>eugenei</i>	1408	UK, 1961
<i>Populus</i> sp.	600	France, 1964

Host	CFBP no.	Origin and year of isolation
<i>Prunus amygdalus</i>	1577	France, 1974
<i>Prunus armeniaca</i>	1578	France, 1974
<i>Prunus armeniaca</i>	1588	Turkey, 1973
<i>Prunus avium</i>	1680	Hungary, 1958
<i>Prunus avium</i>	2006	New Zealand, 1974
<i>Prunus cerasus</i>	1543	France, 1974
<i>Prunus cerasus</i>	2334	France, 1981
<i>Prunus domestica</i>	692	France, 1965
<i>Prunus persica</i>	1582	France, 1969
<i>Prunus persica</i>	1584	France, 1974
<i>Prunus triloba</i>	R10.1 (*)	France, 1982
<i>Prunus triloba</i>	S9.6 (*)	France, 1983
<i>Pyrus communis</i>	11	France, 1957
<i>Pyrus communis</i>	311	France, 1962
<i>Pyrus communis</i>	1391	UK, 1959
<i>Rosa</i> sp.	1608	France, 1975
<i>Rubus idaeus</i>	1586	France, 1974
<i>Salix matsudana</i> var. <i>tortuosa</i>	1692	UK, 1967
<i>Salix</i> sp.	1771	New Zealand, 1968
<i>Sorghum vulgare</i> var. <i>sudanense</i>	1684	Hungary, 1967
<i>Syringa vulgaris</i>	775	France, 1965
<i>Syringa vulgaris</i>	1392	UK, 1950
<i>Thuja</i> sp.	2575	France, 1979
<i>Triticum aestivum</i>	1767	USA, 1972
<i>Vicia villosa</i>	1763	USA, 1975
<i>Zea mays</i>	1685	Yugoslavia, 1965
<i>Zea mays</i>	1894	France, 1978

CFBP = Collection Française de Bactéries Phytopathogènes, INRA, Angers, France.

(*) Laboratory Collection of L. Gardan, INRA, Angers, France.

— 53 type strains of pathovars of *P. syringae* and other phytopathogenic fluorescent *Pseudomonas* species (table II).

Biochemical and physiological tests

Presence of oxidase, fluorescent pigment production, presence of gelatinase, production of levan, acid production from sucrose, sorbitol, erythritol, mannitol, reduction of nitrate, lipolysis of Tween-80, hydrolysis of aesculin, hypersensitive reaction on tobacco leaves and presence of arginine dihydrolase were tested according to Lelliott *et al.* (1966). Pectolytic activity was tested by the method of Prunier and Kaiser (1961) and on pectate gel at pH 5 and 8 (Hildebrand, 1971). Utilization of DL-lactate, L(+)tartrate, D(-)tartrate was tested on basal medium (NH₄H₂PO₄, 1 g; KCl, 0.2 g; MgSO₄·2H₂O, 0.2 g; agar, 3 g; bromothymol blue, 0.08 g; distilled water, 1000 ml; pH 7.2) plus each organic acid, 0.1 w/v. Presence of DNase was tested on DNAgar (Diagnostic Pasteur).

Assimilation of 49 carbohydrates, 49 organic acids and 49 amino acids was studied with API-50-CH, -50-AO and -50-AA galleries (Api System, France) which were incubated at 24°C; growth was recorded after 6 days.

Numerical taxonomy

A total of 167 characters were studied. Logical coding 1/0 was used. The distant matrix was calculated using the Jaccard coefficient (Sneath and Sokal, 1973). Cluster analysis was done using unweighted pair group method with averages (UPGMA) (Sneath and Sokal, 1973).

A numerical identification program calculated the following criteria: theoretical frequency of emergence of a phenotype within the species (product of positivity percentages of the different phenons), which reflected dissimilarity of the phenotype, and identification percentage (theoretical frequency of emergence of the phenotype for the species under the sum of emergence frequencies (Lapage *et al.*, 1973; Bollet

Table II. Origins of strains of phytopathogenic *Pseudomonas* tested.

Name	CFBP no.	Host plant, origin and year of isolation
<i>P.s. pv. aceris</i>	2339 ^T	<i>Acer</i> sp., USA, year unknown
<i>P.s. pv. aesculi</i>	289 ^T	<i>Aesculus indica</i> , Indi, 1980
<i>P.s. pv. anthrini</i>	1620 ^T	<i>Anthriscum majus</i> , UK, 1965
<i>P.s. pv. apii</i>	2103 ^T	<i>Apium graveolens</i> var. <i>dulce</i> , USA, 1942
<i>P.s. pv. aptata</i>	1617 ^T	<i>Beta vulgaris</i> , USA, 1959
<i>P.s. pv. atrofaciens</i>	2213 ^T	<i>Triticum aestivum</i> , New Zealand, 1968
<i>P.s. pv. atropurpurea</i>	2340 ^T	<i>Lolium multiflorum</i> , Japan, 1967
<i>P.s. pv. berberidis</i>	1727 ^T	<i>Berberidis</i> sp., New Zealand, 1972
<i>P.s. pv. cannabina</i>	2341 ^T	<i>Cannabis sativa</i> , Hungary, 1957
<i>P.s. pv. ciccaronei</i>	2342 ^T	<i>Ceratonia siliqua</i> , Italy, 1969
<i>P.s. pv. coronafaciens</i>	2216 ^T	<i>Avena sativa</i> , UK, 1958
<i>P.s. pv. delphinii</i>	2215 ^T	<i>Delphinium</i> sp., New Zealand, 1957
<i>P.s. pv. dysoxyl</i>	2356 ^T	<i>Dysoxylum spectabile</i> , New Zealand, 1949
<i>P.s. pv. eriobotryae</i>	2343 ^T	<i>Eriobotrya japonica</i> , USA, 1970
<i>P.s. pv. glycinea</i>	2214 ^T	<i>Glycine max</i> , New Zealand, 1968
<i>P.s. pv. helianthi</i>	2067 ^T	<i>Helianthus annuus</i> , Mexico, 1972
<i>P.s. pv. hibisci</i>	2895 ^T	<i>Hibiscus japonica</i> , USA, 1984
<i>P.s. pv. japonica</i>	2896 ^T	<i>Hordeum vulgare</i> , Japan, 1951
<i>P.s. pv. lachrymans</i>	2440 ^T	<i>Cucumis sativus</i> , USA, 1935
<i>P.s. pv. lapsa</i>	1731 ^T	<i>Triticum aestivum</i> (hybrid), place unknown, 1968
<i>P.s. pv. maculicola</i>	1657 ^T	<i>Brassica oleracea</i> , New Zealand, 1965
<i>P.s. pv. mellea</i>	2344 ^T	<i>Nicotiana tabacum</i> , Japan, 1968
<i>P.s. pv. mori</i>	1642 ^T	<i>Morus alba</i> , Hungary, 1958
<i>P.s. pv. mors-prunorum</i>	2351 ^T	<i>Prunus domestica</i> , USA, year unknown
<i>P.s. pv. myricae</i>	2897 ^T	<i>Myrica rubra</i> , Japan, 1978
<i>P.s. pv. panici</i>	2345 ^T	<i>Panicum</i> sp., USA, 1963
<i>P.s. pv. papulans</i>	1754 ^T	<i>Malus sylvestris</i> , Canada, 1973
<i>P.s. pv. passiflorae</i>	2346 ^T	<i>Passiflora edulis</i> , New Zealand, 1962
<i>P.s. pv. persicae</i>	1573 ^T	<i>Prunus persica</i> , France, 1974
<i>P.s. pv. persicae</i>	W 24-1	<i>Prunus persica</i> , New Zealand, 1980
<i>P.s. pv. persicae</i>	W 24-2	<i>Prunus persica</i> , New Zealand, 1980
<i>P.s. pv. persicae</i>	W 24-3	<i>Prunus persica</i> , New Zealand, 1980
<i>P.s. pv. phaseolicola</i> (race 1)	1390 ^T	<i>Phaseolus vulgaris</i> , Canada, 1949
<i>P.s. pv. philadelphi</i>	2898 ^T	<i>Philadelphus coronarius</i> , UK, 1982
<i>P.s. pv. photinae</i>	2899 ^T	<i>Photinia glabra</i> , Japan, 1983
<i>P.s. pv. pisi</i>	2105 ^T	<i>Pisum sativum</i> , New Zealand, 1969
<i>P.s. pv. porri</i>	1908 ^T	<i>Allium porrum</i> , France, 1978
<i>P.s. pv. primulae</i>	1660 ^T	<i>Primulae</i> sp., USA, 1939
<i>P.s. pv. ribicola</i>	2348 ^T	<i>Ribes aureum</i> , place unknown, 1946
<i>P.s. pv. savastanoi</i>	1670 ^T	<i>Olea europaea</i> , Yugoslavia, 1959
<i>P.s. pv. sesami</i>	1671 ^T	<i>Sesamum indicum</i> , Yugoslavia, 1961
<i>P.s. pv. striafaciens</i>	1674 ^T	<i>Avena sativa</i> , place unknown, 1966
<i>P.s. pv. syringae</i>	1392 ^T	<i>Syringa vulgaris</i> , UK, 1950
<i>P.s. pv. tabaci</i>	2106 ^T	<i>Nicotiana tabacum</i> , Hungary, 1959
<i>P.s. pv. tagetis</i>	1694 ^T	<i>Tagetes erecta</i> , Rhodesia, 1972
<i>P.s. pv. theae</i>	2353 ^T	<i>Theae sinensis</i> , Japan, 1970
<i>P.s. pv. tomato</i>	2212 ^T	<i>Lycopersicum esculentum</i> , UK, 1960
<i>P.s. pv. ulmi</i>	1407 ^T	<i>Ulmus</i> sp., Yugoslavia, 1958
<i>P.s. pv. viburni</i>	1702 ^T	<i>Viburnum</i> sp., unknown
<i>Pseudomonas amygdali</i>	W 28-1 ^T	<i>Prunus amygdalus</i> , Greece, 1974
<i>Pseudomonas amygdali</i>	2354	<i>Prunus amygdalus</i> , Greece, 1974
<i>Pseudomonas cichorii</i>	2101 ^T	<i>Cichorium endiva</i> , Germany, 1929
<i>Pseudomonas viridiflava</i>	2107 ^T	<i>Phaseolus</i> sp., Switzerland, 1927

CFBP = Collection Française de Bactéries Phytopathogènes, INRA, Angers, France. T = type strain.

et al., 1988). Thanks to this method, it was possible to assign an isolated phenotype to a phenon.

The amount of information for each test was measured by calculating the diagnostic ability coefficient (DAC) (Descamp and Véron, 1981).

RESULTS

The dendrogram displaying the distance relationship among the 161 strains is shown in figure 1.

At distance level 0.158, 14 phenons and 32 isolated strains were observed. The number, the name and the origin of the strains clustered in the different phenons are given in table III.

Phenon 14 included 93 out of 108 strains of pathovar *syringae*. Among these 93 strains, 48 were isolated from cherry laurel and 45 from 31 different hosts. Phenon 14 also included 6 other pathovars: *atrofaciens*, *dysoxyli*, *aptata*, *panici*, *populans* and *aceris*.

Fifteen other strains of pathovar *syringae* were distributed either among 5 other phenons (8, 9, 11, 12 and 13) or isolated strains (CFBP S433, 311, U21-1, 1787, 1574 and 1701). Phenon 9 also included pathovars *tomato*, *maculicola* and *apii*. Phenon 11 also included pathovars *mellea* and *tabaci*.

Other pathovars of *P. syringae* and *P. amygdali* were distributed in phenons (1, 2, 3, 4, 6, 7 and 10).

The type strain of *P. syringae* (two replicates of CFBP 1392) constituted phenon 5 and clustered away from the other strains of pathovar *syringae*.

The distance between the above mentioned pathovars of *P. syringae* and the last 26 pathovars of *P. syringae* was very large.

A numerical identification program linked 2 strains of pathovar *persicae* (CFBP 1573 and W24-3) to the other strains of phenon 2.

We selected 26 biochemical tests with DAC ranging from 0.669 to 0.205, which differentiated the 14 phenons from each other (table IV). However, phenons 13 and 14, which were closely related, were not differentiated by biochemical tests.

DISCUSSION

In this study, 93 of 108 strains of pathovar *syringae* were in phenon 14 with pathovars *atrofaciens*, *dysoxyli*, *aptata*, *panici*, *populans* and *aceris*.

Table III. Number and identity of strains clustered in the 14 phenons.

Phenon	No. of strains	Name of pathovars
1	2	pv. <i>amygdali</i>
2	2	pv. <i>persicae</i>
3	2	pv. <i>philadelphia</i> , pv. <i>berberidis</i>
4	3	pv. <i>photiniae</i> , pv. <i>myricae</i> , pv. <i>aesculi</i>
5	2	pv. <i>syringae</i>
6	2	pv. <i>coronafaciens</i> , pv. <i>striaefaciens</i>
7	2	pv. <i>passiflorae</i> , pv. <i>delphinii</i>
8	2	pv. <i>syringae</i>
9	7	pv. <i>syringae</i> (4 strains) + pv. <i>tomato</i> , pv. <i>maculicola</i> , pv. <i>apii</i>
10	2	pv. <i>ribicola</i> , pv. <i>primulae</i>
11	3	pv. <i>syringae</i> , pv. <i>mellea</i> , pv. <i>tabaci</i>
12	2	pv. <i>syringae</i>
13	2	pv. <i>syringae</i>
14	97	pv. <i>syringae</i> (91 strains) + pv. <i>atrofaciens</i> , pv. <i>dysoxyli</i> , pv. <i>aptata</i> , pv. <i>panici</i> , pv. <i>populans</i> , pv. <i>aceris</i>

Fig. 1. Dendrogram of similarity based on the distance of 161 strains of *P.s. pv. syringae*, pathovars of *P. syringae* and phytopathogenic fluorescent *Pseudomonas*.

■ = *P.s. pv. syringae* from cherry laurel; ▲ = *P.s. pv. syringae* from various hosts.

The numerical taxonomy of Sands *et al.* (1970) and the DNA relatedness study of Pecknold and Grogan (1973) also grouped pathovar *syringae* with pathovars *aptata* and *panici*. However, pathovars *pisi* and *tabaci* (synonym *angulata*) which clustered with pv. *syringae* in the study of Sands *et al.* (1970) were unclustered and in phenon 11 (respectively) in this study. Pathovar *pisi* was highly related to pv. *syringae* in DNA pairing (Pecknold and Grogan, 1973). However, neither Sands *et al.* (1970) nor Pecknold and Grogan (1973) included the reference strains of pathovar *pisi* or *tabaci*.

The type strain of *P.s.* pv. *syringae* (CFBP 1392) was distant from phenon 14 and thus is not truly representative of pv. *syringae*. The present biochemical definition of pv. *syringae* should be amended, since 15 strains identified

as *P.s.* pv. *syringae*, according to this definition, were scattered along the dendrogram shown in figure 1. However, some of these strains could have been misidentified. Biochemical characters listed in table IV could be useful for the identification of the phenons delineated in this study.

Phenon 14 appears to be correspond to pv. *syringae*. Our study suggests that it is not accurate to maintain the six pathovars (*atrofaciens*, *dysoxyl*, *aptata*, *panici*, *papulans* and *aceris*) clustered in phenon 14 as distinct pathovars.

Our study confirms that phenons 1 and 2 were, respectively, equivalent to *P. amygdali* and pv. *persicae*. It would be necessary to confirm this by using more strains.

"The term of pathovar referred to a strain or set of strains with the same or similar charac-

Table IV. Biochemical characteristics of the 14 phenons delineated in this study.

Test	Characteristics of phenons													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
— Fluorescence	+	—	+	+	+	d	+	+	+	—	+	+	+	+
— Hydrolysis of:														
Esculin	+	—	+	—	—	+	+	+	+	—	—	d	+	+
Gelatin	+	—	+	d	d	—	d	—	—	—	—	—	—	d
Pectate pH 5	—	—	—	—	—	—	d	+	—	+	—	—	—	—
— Utilization of:														
DL-lactate	+	—	+	—	—	—	—	—	+	—	—	d	+	+
Meso-erythritol	+	—	+	—	—	+	+	+	+	—	—	—	+	+
DL-5-hydroxybutyrate	+	—	+	+	+	—	+	—	+	—	—	—	+	+
Caprylate	+	—	+	+	+	—	+	+	+	—	—	+	+	+
Betaine	+	—	+	+	+	+	+	+	+	—	—	+	+	+
D(-)tartrate	—	+	—	+	+	—	d	—	+	—	+	—	—	d
n-Caproate	+	—	—	—	—	—	+	—	—	—	—	—	+	d
L-leucine	+	—	+	+	+	+	+	+	+	—	d	d	—	+
Caprate	+	—	+	+	+	d	+	+	+	—	—	+	+	+
D-glucosamine	+	—	+	+	—	—	d	—	d	—	—	—	+	d
Meso-tartrate	+	—	—	+	+	+	+	+	+	—	+	+	+	+
Sarcosine	+	—	+	—	d	+	+	—	—	—	—	d	—	d
Pelargonate	+	—	+	+	d	d	+	d	+	—	d	+	+	+
Inositol	+	—	+	+	+	+	+	+	+	—	+	+	+	+
DL-5-aminovalerate	+	—	+	+	d	d	+	—	d	—	—	+	d	d
L-histidine	+	—	—	+	+	+	+	d	+	d	+	+	+	+
L(+)tartrate	—	—	—	—	—	—	d	—	—	—	—	+	—	—
Heptanoate	+	+	+	+	d	d	+	d	+	—	+	+	+	+
L-arginine	+	+	—	d	d	d	+	d	d	—	+	+	+	+
Trigonelline	+	+	+	+	d	—	+	d	+	—	+	+	+	+
Number of strains	2	2	2	2	7	2	3	2	2	2	2	3	2	97

+ = 90 % or more of strains are positive; — = 90 % or more of strains are negative; d = 10 to 89 % of strains are positive.

teristics, differentiated at infrasubspecific level from other strains of the same species or subspecies on the basis of distinctive pathogenicity to one or more plants" (Dye *et al.*, 1980). The term pathovar was proposed by Young *et al.* (1978) at the demand of plant pathologists, for phytopathogenic bacteria which are, or are believed to be, highly specific for their hosts. Like Vauterin *et al.* (1990), we think that this special-purpose classification does not meet the requirements for naming taxa according to the *International Code of Nomenclature of Bacteria* (Lapage *et al.*, 1975). Indeed, this nomenclature is open to criticism.

Phytopathogenicity is only one of the phenotypic properties of plant pathogenic bacteria. However, directly or indirectly, it probably represents a large number of genes in the genome of bacteria. Some pathogenic bacteria have a very broad host-range because they can attack a variety of host plant species, whereas others may attack a single host species. Thus, it is sometimes very difficult to prove that a bacteria is pathogenic on one or several host plants. For the pathovars of *P. syringae*, we have no comprehensive data on cross-pathogenicity.

On the other hand, pathogenicity is not correlated with phenotypic properties. We have observed that only 30 of 50 strains of pv. *syringae* isolated from cherry laurel and belonging to phenon 14 were in fact pathogenic for cherry laurel, whereas 10 of 31 strains of pv. *syringae* isolated from other hosts and belonging to phenon 14 were pathogenic for cherry laurel (unpublished data).

DNA relatedness is the best indication of genospecies. It is currently generally accepted that DNA-DNA hybridization will be essential to clarify the taxonomy of plant pathogenic bacteria, and especially fluorescent *Pseudomonas*. Such work is now in progress in our laboratory.

Acknowledgements

We would like to thank Drs. Francine and P.A.D. Grimont for helpful discussion and for critical reading of the manuscript.

Étude de l'hétérogénéité phénotypique de *Pseudomonas* pv. *syringae* van Hall

L'étude des caractères phénotypiques de 108 souches de *Pseudomonas syringae* pv. *syringae* isolées de laurier palme (50) et d'hôtes variés (58) en comparaison avec les souches types des pathovars de *P. syringae*, a montré que 91 souches sur 108 s'agrégent dans le phénon 14 avec 6 autres pathovars de *P. syringae* (*aceris*, *atrofaciens*, *aptata*, *dysoxyli*, *panici* et *papulans*). On pourrait donc considérer les souches du phénon 14 comme appartenant à l'espèce *P.s.* pv. *syringae*.

P. amygdali et *P.s.* pv. *persicae* forment des phénons distincts de *P.s.* pv. *syringae*. Un certain nombre d'autres pathovars *syringae* se regroupent dans plusieurs mêmes phénons; les autres souches sont considérées comme des phénotypes isolés.

Les 14 phénons sont différenciables par les caractères biochimiques.

Mots-clés: *Pseudomonas syringae*; Phénotypage, Pathovars, Taxonomie numérique.

References

- Bradbury, J.F. (1986), *Pseudomonas*, in "Guide to plant pathogenic bacteria" (pp. 110-185). CAB International Mycological Institute, Kew, U.K.
- Bollet, C., Gullian, C., Chaudet, H., Fichet, B., Aragon, A. & de Micco, P. (1988), Identification des principales espèces du genre *Serratia* par une galerie simplifiée. *Ann. Inst. Pasteur/Microbiol.*, 139, 337-349.
- Descamp, P. & Véron, M. (1981), Une méthode de choix des caractères d'identification basée sur le théorème de Bayes et la mesure de l'information. *Ann. Microbiol. (Inst. Pasteur)*, 132B, 157-170.
- Dye, D.W., Bradbury, J.F., Goto, M., Hayward, A.C., Lelliott, R.A. & Schroth, M.N. (1980), International standards for naming pathovars of phytopathogenic bacteria and a list of pathovar names and pathotype strains. *Rev. Plant Path.*, 59, 153-168.
- Hildebrand, D.C. (1971), Pectate and pectin gels for differentiation of *Pseudomonas* sp. and other bacterial plant pathogens. *Phytopathology*, 61, 1430-1436.
- Hildebrand, D.C. & Schroth, M.N. (1972), Identification of the fluorescent *Pseudomonads*, in "Proceedings of 3rd International Conference on Plant Pathogenic Bacteria, Wageningen, 14-21 april 1971, 281-287.
- Lapage, S.P., Bascomb, S., Wilcox, W.R. & Curtis, M.A. (1973), Identification of bacteria by computer: general aspects and perspectives. *J. gen. Microbiol.*, 77, 273-290.
- Lapage, S.P., Sneath, P.H.A., Lessel, E.F., Skerman, U.B.D., Seelinger, H.P.R. & Clark, W.A. (1975),

- International code of nomenclature of bacteria. 1975 Revision. American Society for Microbiology, Washington, D.C.
- Lelliott, R.A., Billing, E. & Hayward, A.C. (1966), A determinative scheme for the fluorescent plant pathogenic *Pseudomonads*. *J. appl. Bact.*, 29, 470-489.
- Misaghi, I. & Grogan, R.G. (1969), Nutritional and biochemical comparisons of plant pathogenic and saprophytic fluorescent *Pseudomonads*. *Phytopathology*, 59, 1436-1450.
- Pecknold, P.C. & Grogan, R.G. (1973), Deoxyribonucleic and homology groups among phytopathogenic *Pseudomonas* species. *Int. J. syst. Bact.*, 23, 111-121.
- Prunier, J.P. & Kaiser, P. (1961), Etude de l'activité pectinolytique chez des bactéries phytopathogènes et saprophytes des plantes. — I. Recherche des enzymes pectinolytiques. *Ann. Epiphyties*, 15, 205-219.
- Sands, D.C., Schroth, M.N. & Hildebrand, D.C. (1970), Taxonomy of phytopathogenic *Pseudomonads*. *J. Bact.*, 9-23.
- Sneath, P.H.A. & Sokal, R.R. (1973), Numerical taxonomy. The principles and practice of numerical classification. Freeman & Co., San Francisco.
- Stanier, R.Y., Palleroni, N.J. & Doudoroff, M. (1970), The aerobic *Pseudomonads*: a taxonomic study. *J. gen. Microbiol.*, 43, 159-271.
- Vauterin, L., Swings, J., Kersters, K., Gillis, M., Mew, T.M., Schroth, M.N., Palleroni, N.J., Hildebrand, D.C., Stead, D.E., Civerolo, E.L., Hayward, A.C., Maraitre, H., Stall, R.E., Vidaver, A.K. & Bradbury, J.F. (1990), Toward an improved taxonomy of *Xanthomonas*. *Int. J. syst. Bact.*, 40, 312-316.
- Young, J.M., Dye, D.W., Bradbury, J.F., Panagopoulos, C.G. & Robbs, C.F. (1978), A proposed nomenclature and classification for plant pathogenic bacteria. *N.Z. J. Agric. Res.*, 21, 153-177.