II 식물 병원성 *Corynebacterium, Agrobacterium, Psuedomonas*의 분류

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과거의 모든 세균 분류는 세균의 형태에 의존하였다. 아직도 세균의 형태는 분류에서 기본이 되나 최근 새로 개발된 많은 분류 방법에 의해 다각적인 test를 하여야만 새로운 분류가 인정받게 된다. Polyphasic test 에는 생리. 생태. 생화학적 분류 방법을 기본으로 하여, DNA-DNA hybridization, GC ratio, DNA-rRNA hybridization등의 분자 생물학적 방법, 세포벽의 성분 분석, 지질 분석, 단백질 분석 등의 방법이 포함되며, 이러한 polyphasic test에 의하여 최근 많은 식물 병원 세균이 재분류되고 있다. 본 review에서는 기본의 세균 분류와 비교, 최근에 재분류된 식물 병원세균의 소개에 초점을 맞추었다.

KINGDOM PROKAROYOTAE
- BACTERIA-have cell membrane and cell wall
  - Division: FIRMICUTES-Gram positive bacteria
    - Class: THALLOBACTERIA-Branching bacteria
      - Family: No family classification in Firmicutes
        - Genus: *Arthrobacter*
        - Genus: *Clavibacter*
        - Genus: *Corynebacterium*
        - Genus: *Curtobacteirum*
        - Genus: *Rhodococcus*
        - Genus: *Rathayibacter*
  - Division: GRACILICUTES-Gram negative bacteria
    - Class: PROTEOBACTERIA-mostly single celled bacteria
      - Alpha subclass
        - Family: Rhizobiaceae
          - Genus: *Agrobacterium*
        - Family: Not clssified
          - Genus: *Rhizomonas*
      - Beta subclass
        - Family: Comamonadaceae
          - Genus: *Acidovorax*
          - Genus: *Comamonas*
          - Genus: *Hydrogenophaga*
          - Genus: *Burkholderia*
          - Genus: *Ralstonia*
          - Genus: *Xylophilus*

Gamma subclass
Family: Pseudomonadaceae
Genus: *Pseudomonas*

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1. Coryneform 식물 병원 세균의 재분류

1934년 Jensen이 알팔파 시들음병과 토마토 궤양병을 일으키는 세균을 각각 *Corynebacterium insidiosum*과 *C. michiganense*로 분류한 이래 많은 spore를 형성하지 않는 그램 양성 간균이 genus *Corynebacteirum*에 포함되었다. 이러한 분류에는 많은 문제점이 있었으며, 이러한 문제점을 해결하기 위하여 Coryneform bacteria의 재분류가 시도되었다. 생리. 생태. 생화학적 분류 방법을 기본으로 하고 Coryneform bacteria의 독특한 세포벽과 지질의 조성 물질을 이용하여 *Corynebacterium* 은 *Arthrobacter, Clavibacter, Curtobacterium, Rhodococcus,* 그리고 *Rathayibacter*로 재분류되었다. 재분류의 기준에 따라, genus *Corynebacterium* 에 속해있던 식물병원세균은 모두 다른 genus로 재분류되었다.

*** 다음의 대부분의 분류 설명은 1994년 발간된 Bergey's Manual에서 발췌하였다.

(1) GROUP 20 IRREGULAR, NONSPORING GRAM-POSITIVE RODS

○ Genus *Arthrobacter*

Cells in young cultures are irregular rods, 0.8-1.2 X 1.0-8.0 μm, often V-shaped and with clubbed ends, but there are no filaments. As growth proceeds the rods segment into small cocci, 0.6-1.0 μm in diameter, arranged singly, in pairs, and in irregular clumps. This marked rod-coccus growth cycle is characteristic of *Arthrobacter*; stationary phase cultures consist almost entirely of cocci. Gram positive but easily decolorized. The rods of some species are motile. Nonsporing, not acid-fast. Aerobic. Chemoorganotrophic, usually grow on simple media plus biotin, with an oxidative metabolism. Little or no acid and no gas is produced from glucose and other carbohydrates. Catalase positive. The optimum growth temperature is 25-30℃. Widely distributed in the environment, principally in soils.

○ Genus *Clavibacter*

Straight or slightly curved slender rods, 0.4-0.75 x 0.8-2.5 μm, irregular and often wedge- or club-shaped; predominantly arranged singly but often are arranged in pairs in a V configuration and sometimes in palisades. Old cultures are not predominantly coccoid. Gram positive, nonmotile, nonsporing, not acid-fast. Obligate aerobes, require nutritionally rich media

on which they grow slowly. Chemoorganotrophic, metabolism respiratory, producing weak acidity from glucose and some other carbohydrates. The optimum temperature is 20-29℃; seldom grow above 35℃. Some strains produce yellow or blue pigments.. Catalase positive, oxidase negative, nitrate not reduced, indole negative. Obligate parasites of various flowering plants, in which they are pathogenic. Type species: *Clavibacter michiganensis*.

○ Genus *Corynebacterium*

Straight or slightly curved, slender rods have tapered or sometimes clubbed ends and are 0.3-0.8 x 1.5-8.0 μm; Cells are usually arranged singly or in pairs, often in a V formation or in palisades of several parallel cells. Gram positive, though some cells stain unevenly, giving a beaded appearance. Metachromatic granules of polymetaphosphate are commonly formed within the cells. Nonmotile, nonsporing, not acid-fast. Facultative anaerobes, commonly requiring nutritionally rich media such as serum or blood media. on which colonies are usually convex and semiopaque, with a mat surface. Chemoorganotrophs with fermentative metabolism, most species produce acid without gas from glucose and some other carbohydrates. Catalase positive, often reduce nitrate and ellurite. Rarely acidify lactose or raffinose or liquefy gelatin. Primarily obligate parasites of mucous membranes or skin of mammals, but occasionally they are found in other sources; some species are pathogenic for mammals. Type species: *Corynebacterium diphtheriae*

○ Genus *Curtobacterium*

Small, short, irregular rods in young cultures, 0.4-0.6 x 0.6-3.0 μm, become coccoid in old cultures. Arranged singly or sometimes in pairs, often in a V formation; no branching is found. Gram positive, but cells from old cultures are easily decolorized. Generally motile by peritrichous flagella. Nonsporing, not acid-fast. Metachromatic granules are absent. Obligately aerobic, they yield smooth, convex colonies on nutrient agar; the colonies are usually yellow or orange. Chemoorganotrophic, not especially exacting nutritionally. Metabolism is respiratory, yielding small amounts of acid from glucose and some other carbohydrates. Catalase positive. The optimum growth temperature is 25-30℃. Occur on plants, in soil, and in oil brine; *C. flaccumfaciens* is a plant pathogen.

○ Genus *Rathayibacter*

A new genus, *Rathayibacter,* is proposed to accomodate three species of gram-positive, aerobic, coryneform bacteria previously placed in the genus *Clavibacter*, as well as six strains that were isolated from annual cereal grasses, may be responsible for ryegrass toxicity. the properties of members of the genus *Rathayibacter* include coryneform morphology, peptidoglycan based on 2,4-diaminobutyric acid, predominant menaquinones of the MK-10 type, and phosphatidylglycerol and diphosphatidylglycerol as basic polar lipids. The DNA base compositions range from 63 to 72

mol% G+C. The members of the new genus from a phenetic cluster distinct from *Clavibacter* spp. at the level of 70% (simple matching coefficient) and exhibit 7 to 9% DNA-DNA reassociation with strains of *Clavibacter* spp. In contrast to *Clavibacter* spp., most *Rathayibacter* strains are associated with nematodes belonging to the genus *Anguina*. The *Rathayibacter* species differ from species belonging to related genera in the following characteristics: menaquinone and whole-cell sugar compositions, results of lysozyme, sodium dodecyl sulfate test (which indicates differences in cell wall composition), ability to utilize a number carbon sources, resistance to bacteriocins of some *Clavibacter* spp., and other characteristics. The *Rathayibacter* species can be differentiated from each other by following characteristics: presence or absence of xylose and galactose in the cell walls, fatty acid composition, ability to assimilate various sources of carbon and nitrogen, hydrolytic activity, tolerance to 5% NaCl and 0.03% potassium tellurite, susceptibility to iranicin, and absence or presence of plasmids of certain sizes. (Zgurskaya H. I. et al., 1993. Int. J. Syst. Bacteriol. 43:143-149)

Table Characteristics differentiating the species and subspecies of *Clavibacter*[a,b]

Characteristics	*C. iranicus*	*C. michiganensis* subsp. *insidiosus*	*C. michiganensis* subsp. *michiganensis*	*C. michiganensis* subsp. *nebraskensis*	*C. michiganensis* subsp. *sepedonicus*	*C. michiganensis* subsp. *tesselarius*	*C. rathayi*	*C. tritici*	*C. xyli* subsp. *cynodontis*	*C. xyli* subsp. *xyli*
Colonies with:										
Yellow or orange pigment	+	+	+	+	–	+	+	+	+	–
Blue or grey pigment	–	d	d	–	–	–	–	–	–	–
Acid produced from:										
Inulin	–	–	–	–	–	–	–	+	–	–
Mannitol	–	–	–	–	+	+	+	+	+	+
Mannose	+	+	+	+	d	ND	–	+	+	+w
Melezitose	+	–	–	–	–	–	–	–	–	–
Sorbitol	–	–	–	+	+	+	–	–	–	–
Utilization of:										
Acetate	–	–	+	+	+	+	–	+	–	–
Citrate	+	+	+	+	+	+	+	+	+	–
Lactate	–	–	d	+	–	ND	–	–	–	–
Propionate	–	–	–	+w	–	–	–	–	–	–
Succinate	+	–	+	+	+	ND	+	+	–	–
Hydrolysis of:										
Gelatin	–	–	+w	–	–	–	+	–	–	–
Soluble starch	–	–	d	d	d	+	d	–	+	–
Methyl red	–	+	–	d	–	–	d	–	–	–
H_2S from peptone	+	–	+	d	–	–	+	+	–	–

[a] Symbols: +, 90% or more of strains are positive; –, 90% or more of strains are negative; d, 11–89% of strains are positive; w, weak reaction; ND, not determined.

[b] From *Bergey's Manual of Systematic Bacteriology* and Davis et al. (Int. J. Syst. Bacteriol. *34:* 107–117, 1984).

Table. Some diagnostic and differentiating characteristics of the genera *Clavibacter*, *Rathayibacter*, and *Agromyces*

Characteristics	*Clavibacter*	*Rathayibacter*	*Agromyces*
Peptidoglycan based on DAB	+	+	+
Major menaquinone	MK-9	MK-10	MK-12
Cell wall sugars			
Glucose	+	+	(+)
Galactose	+	(+)	+
Mannose	+	+	(+)
Rhamnose	+	+	+
Xylose	-	(+)	(+)
Fucose	(+)	-	(+)
Tyvelose	-	-	(+)
Mycelium	-	-	+
Utilization of:			
Inositol	+	-	(+)
Melibiose	(+)	-	(+)
L-Rhamnose	(+)	-	+
Tagatose	(+)	-	-
Susceptibility to iranicin	+	(+)	-
Lysozyme-SDS test	+	-	+
Usual sources of isolation	plants or plant substrate	plants	soil

Table. Levels of DNA relatedness among *Clavibacter*, *Rathyibacter*, and *Agromyces* strains

Strain	% Relatedness to [H^3]DNA from		
	ICMP 2550	ICMP 2574	VKM Ac1340
Clavibacter michiganensis subsp. *michiganensis* ICMP 2550	100	7	10
Clavibacter michiganensis subsp. *nebraskensis* ICMP 3298	60		
Clavibacter michiganensis subsp. *tessellarius* ICMP 7221	56		
R. rathayi ICMP 2574	9	100	12
R. tritici ICMP 2626	8	43	
R. iranicus ICMP 3496	8	16	
Agromyces ramosus ATCC 25173	7	5	12
Agromyces cerinus subsp. *cerinus* VKM Ac 1340	10	7	100
Agromyces fucosus subsp. *fucosus* VKM Ac 1345			40

(2) GROUP 22 NOCARDIOFORM ACTINOMYCETES

○ Genus *Rhodococcus*

Rods to extensively branched vegetative mycelium may be formed. In all strains the morphogenetic cycle is initiated with the coccus or short rod stage, with different organisms showing a succession of more or less complex morphological stages by which completion of the growth cycle is achieved. Thus, cocci may merely germinate into short rods, form filaments with side projections, show elementary branching, or, in the most differentiated forms, produce extensively branched hyphae. The next generation of cocci or short rods is formed by fragmentation of the rods, filaments, and hyphae. Some strains produce feeble, microscopically visible aerial hyphae, which may be branched, or aerial synnemata consisting of unbranched filaments that coalesce and project upward. Colonies may be rough, smooth, or mucoid and pigmented buff, cream, yellow, orange, or red, although colorless variants do occur. Stain gram positive. Usually partially acid-fast. Aerobic. Do not contain mycobactins. Sensitive to lysozyme. The glycan moiety of the cell wall has N-glycolyl residues. The wall envelope contains mycolic acids with 34 to 52 carbon atoms and up to 3 double bonds and major proportions of straight-chain saturated, unsaturated, and 10-methyl (tuberculostearic)-branched fatty acids. Fatty acid esters released on pyrolysis gas chromatography of mycolic esters have 12 to 18 carbon atoms. Cells contain diphosphatidylglycerol, phosphatidylethanolamine, and phosphatidylinositol dimannosides as major phospholipids. Dihydrogenated menaquinone with eight isoprene units (MK-8(H_2)) form the predominant isoprenolog. Widely distributed but particularly abundant in soil and herbivore dung. Some strains are pathogenic for animals, including human beings.

Table Differential characteristics of phytopathogenic coryneform bacteria.

Pathogen	Mot	Pig.[d]	Growth		Acid Production[a]			Utilization[b]		Hydrolysis[c]	
			CNS	TTC	Ribose	Sorbitol	Inulin	Acetate	Formate	Casein	Esculin
Arthrobacter											
ilicis	+	Y	ND	ND	+	−	−	+	+	+	+
Rathayibacter											
iranicum	−	Y	−	ND	−	−	+	−	−	−	+
rathayi	−	Y	V	ND	−	−	−	−	−	−	+
tritici	−	Y	+	ND	−		+	+	−	−	+
Clavibacter											
michiganensis	subsp.										
insidiosum	−	Y/B	−	+	−	−	−	−	−	−	+
michiganensis	−	Y/V_1	+	+	−	−	−	−	−	−	+
nebraskense	−	O/V_2	+	−	−	+	−	+	−	−	+
sepedonicum	−	W	−	−	−	+	−	+	−	−	+
tessellarius	−	O	+	+	−	+	−	−	−	−	+
xyli subsp.											
cynodontis	−	Y	ND	ND	−	−	−	−	−	−	−
xyli	−	W	ND	ND	−	−	−	−	−	−	−
Curtobacterium											
flaccumfaciens	subsp.										
betae	V	Y	ND	ND	+	+	−	+	−	−	+
flaccumfaciens	V	Y/O/P	ND	ND	+	−	−	+	−	+	+
oortii	+	Y	ND	ND	+	−	−	−	−	+	+
poinsettiae	V	O	ND	ND	+	+	−	+	−	+	+
Rhodoccus											
facians	−	O	ND	ND	+	+	−	+	+	−	−

[a] RSD broth with yeast extract reduced to 0.1 g/l, the test compound (0.5% w/v) replacing glucose, and bromothymol blue (0.008% w/v) blue added.

[b] RSD broth as above, except the test compound at 0.1% w/v and bovine serum albumin omitted from medium when testing *Arthrobacter* and *Curtobacterium* species.

[c] SC medium supplemented with 1% w/v casein or 0.1% esculin and 0.05% ferric citrate. Clearing of medium with casein or the development of a brown color in the medium with esculin indicates hydrolysis.

[d] NBY agar used for all pathogens, except SC agar used for *C. xyli* subspecies. Y, yellow; B, blue; O, orange; W, white or colorless; P, purple; V_1, various pigments (occasional variants are pink, red, orange, and white or colorless); V_2, occasional variants are yellow; ND, not done. Colors refer to those of nondiffusable pigments, except as noted. The blue pigment is intracellular indigoiodine granules sometimes produced in addition to the yellow pigment. The purple pigment is extracellular and occasionally found.

■ Recipes for selective media.

1) Medium CNS

To make 0.5 liter, add the following to a 1 liter flask:

Nutrient Broth	8.0 g
Yeast extract	2.0 g
Potassium phosphate, dibasic	2.0 g
Potassium phosphate, monobasic	0.5 g
Lithium chloride*	5.0 g
Agar	6.5 g
Double distilled water	480.0 ml

Autoclave together (after autoclaving the pH is 6.9), cool to about 50℃, then add the following ingredients:

Cycloheximide	0.020 g (or 2.0 ml of 1g/100ml dH_20. Store refrigerated.)
Nalidixic acid	0.0125 g (or 1.25 ml of 0.1 g in a solution of 7 ml dH_2O + 3 ml 1N NaOH. Store refrigerated.)
Polymixin B sulfate** (8000 USP units/mg)	0.016 g (or 1.6 ml of 1 g/100ml dH_2O. Store refrigerated.)
Daconil 2787-F (530 mg chlorothalonil/ml)	0.00048 g (or 0.03 ml of 1.2 ml/38.8 ml dH_2O. Store at RT.
Glucose	2.5 g (or 25.0 ml of 10% w/v sterile solution)
Magnesium sulfate, anhydrous	0.062 g (or 0.05 ml of 1.0M sterile solution.)

※ The LiCl is transiently toxic to freshly isolated *Clavibacter michiganense* subsp. *nebraskense* hence it can be either imitted from the medium or plating of plant suspensions (in buffer containing sodium) can be delayed for 1-2 hours.

※※ Polymixin B sulfate preparations can vary, hence the medium sould be pretested to determine growth of single colonies.

2) SC medium

Phytone peptone or Soytone	8.0 g
Hemin chloride	15.0 ml (of 0.1% w/v in 0.05 N NaOH)
KH_2PO_4	1.5 g
$K_2HPO_4 \cdot 3H_2O$	0.5 g
$MgSO_4 \cdot 7H_2O$	0.2 g
Corn meal agar	17.0 g
Glucose	1.0 ml (of 50% w/v aqueous solution)
L-Cystein (free base)	10.0 ml (of 10% w/v aqueous solution)
Bovine serum albumin fraction V	10.0 ml (of 20% w/v aqueous solution)

3) TTC medium

Glucose	10.0 g
Peptone	10.0 g
Casamino acids (casein hydrolysate)	1.0 mg
Agar	18.0 g

Autoclave and add 1.0 ml of a 1% (w/v) aqueous solution of 2, 3, 5, triphenyl tetrazolium chloride (autoclaved separately) to each 200 ml portion.

4) RSD broth medium

Yeast extract	1.0 g
$(NH_4)_2HPO_4$	1.0 g
Bovine hemin chloride	15.0 ml (of 0.1% w/v in 0.05 N NaOH)
L-Cystein (free base)	1.0 g
Bromothymol blue	1.0 ml (of 0.8% w/v aqueous solution)
$MgSO_4 \cdot 7H_2O$	0.2 g
KCl	0.2 g
Glucose	2.0 g
Bovine serum albumin fraction V	10.0 ml (of 20% w/v aqueous solution)

Ingredients are added and dissolved in the order given. All ingredients, except for bovine serum albumin, are mixed and the pH adjusted to 7.2 with 0.1 N NaOH or HCl before autoclaving. The bovine serum albumin is filter strilized and added to the medium at room temperature. The final pH should be 6.7.

2. *Agrobacterium*의 분류

Genus *Agrobacterium*은 family Rhizobiaceae에 속한다. 최근 biovar 3을 새로운 species, *A. vitis*로 재분류하였다.

○ Genus *Agrobacterium*

The rods are 0.6-1.0 x 1.5-3.0 μm and occur singly or in pairs. they are non-spore-forming and gram negative. Motility occurs by 1 to 6 peritrichous flagella. Aerobic, possessing a respiratory type of metabolism with oxygen as the terminal electron acceptor. Some strains are capable of anaerobic respiration in the presence of nitrate. Most strains are able to grow under reduced oxygen tensions in plant tissues. Optimum temperature is 25-28℃. Colonies are usually convex, circular, smooth, and nonpigmented to light beige. Growth on carbohydrate-containing media is usually accompanied by copious extracellular polysaccharide slime. Catalase positive and usually oxidase and urease positive. 3-ketoglucosides are produced by the majority of strains belonging to *A. tumefaciens* biovar 1 and *A. radiobacter* biovar 1. Chemoorganotrophs, utilizing a wide range of carbohydrates, salts of organic acids, and amino acids as carbon sources but not cellulose, starch, agar of glucose, D-galactose, and other carbohydrates. Ammonium salts and nitrates can serve as nitrogen sources for strains of some species and biovars; others require amino acids and additional growth factors. With the exception of *A. radiobacter*, members of this genus invade the crown, roots, and stems of a great variety of dicotyledonous and some gymnospermous plants via wounds, causing the transformation of the plant cells into autonomously proliferating tumor cells. The induced plant diseases are commonly known as crown gall, hairy root, and cane gall. Some strains possess a wide host range, whereas other possess a very limited host range. The tumors are self-proliferating and graftable. The tumor induction by *Agrobacterum* is correlated with the presence of a large tumor-inducing plasmid (Ti-plasmid) in the bacterial cells. Agrobacteria are soil inhabitants. Oncogenic strains occur mainly in soils previously contaminated with diseased plant material. Some nononcogenic *Agrobacterium* strains have been isolated from human clinical specimens.

The species nomenclature of *Agrobacterium* is based on phytopathogenic behavior. Those strains causing crown gall are placed in *Agrobacterium tumefaciens,* those causing hairy root in *A. rhizogenes,* those causing cane gall on *Rubus* spp. in *A. rubi* and nonpathogens in *A. radiobacter*. A major problem with this nomenclature is the fact that genes for pathogenicity are carried on large Ti (tumor inducing) or Ri (root inducing) plasmids which can be lost or transferred to a nonpathogenic strain, which in turn becomes pathogenic. Furthermore no correlation exists between the current nomenclature and the taxonomic structure based on morphological, physiological, and genotypical traits of the genus. Chromosomal DNA and

comparison of electrophoretic protein patterns show that the genus *Agrobacterium* consists of at least three taxonomic groups. These groups were proposed to be rased to the species level. Only one is officially accepted.

Table. Differential characteristics of the species of the genus *Agrobacterium*[a]

Characteristic	*A. radiobacter* biovar 1	*A. radiobacter* biovar 2	*A. rhizogenes*[b] biovar 2	*A. rubi*[c]	*A. tumefaciens* biovar 1	*A. tumefaciens* biovar 2	*A. tumefaciens* biovar 3
Growth:							
at 35°C:	+	−	−	d	+	−	d
On selective medium of Scroth et al.[d]	+	−	−		+	−	−
On selective medium of New and Kerr[e]	−	+	+		−	+	−
In presence of 2% NaCl	+	−[f]	−		+	−[f]	+
3-Ketolactose produced	+	−	−	−	+	−	−[f]
Acidic reaction produced from:							
meso-Erythritol	−	+	+	+	−	+	−[f]
Melezitose					+	−	−[f]
Ethanol	+	−	−	−	+[f]	−[f]	−[f]
Alkaline reaction produced from:							
Sodium malonate	−	+	+	+	−	+	+
Sodium L-tartrate					d	+	+
Sodium propionate					d	−	−
Simmons' citrate with 0.0005% yeast extract	−	+	+	−	−	+	
Reaction in litmus milk:[g]							
Alkaline	+	−	−	+	+	−	+
Acidic	−	+	+	−	−	+	
Formation of pellicle in ferric ammonium citrate solution	+	−	−	−	+	−	d
Growth factor requirements:							
Biotin and/or glutamic acid	−	+[f]	+[f]		−	+[f]	
L-Glutamic acid and yeast extract	−	−	−	+	−	+	
Phytopathogenicity:							
Tumors produced on wounded stems of e.g., tomato plants, *Helianthus annuis, Nicotiana tabacum* and/or on discs of *Daucus carota*	−	−	+	+	+	+	d[h]
Roots produced on discs of *Daucus carota*	−	−	+	−	−	−	−

[a] Symbols: see standard definitions.

[b] The majority of the investigated strains belong to biovar 2. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, 248-252, for further information.

[c] Only the following three strains are considered to belong to *A. rubi:* ATCC 13334, 13335, and Braun EU6. Thus only these strains should be used as reference strains for comparison with an unidentified isolate.

[d] Scroth et al., Phytopathology *55*: 645-647, 1965.

[e] New and Kerr, J. Appl. Bacteriol. *34*: 233-236, 1971.

[f] Some strains have been reported to give reactions different from those indicated. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 252, for further information.

[g] An alkaline reaction in litmus milk is frequently accompanied by a brown discoloration; and acid reaction (pink color) is frequently accompanied by a clot formation.

[h] Biovar 3 strains have been isolated mainly from grapevines. The majority of these isolates display a very limited host range. For such strains phytopathogenicity can only be demonstrated on young shoots of grapevines.

○ ***Agrobacterium vitis* sp.nov. *Agrobacterium vitis*** (L. n. *Vitis*, generic name of grapevine)

Some *Agrobacterium* isolates from grapevines that were previously characterized as biovar 3 strains were reclassified into new species *Agrobacterium vitis*. (Ophel Kathy and Allen Jerr. 1990. *Agrobacterium vitis* sp. nov. for strains of *Agrobacterium* biovar 3 from grapevines. Int. J. Syst. Bacteriol. 40:236-241)

Table. Diagnostic characteristics used to identify *Agrobacterium* groups

Characteristic	*A. tumefaciens* NCPPB 2437	*A. radiobacter* ATCC 19358	*A. rhizogenes* ATCC 11325	*A. rubi* TR3	*A. vitis* (biovar 3)
Growth on:					
Selective medium 1	+	+	−	−	−
Selective medium 2	−	−	+	−	−
Selective medium 3	−	−	−	−	+
Growth factor requirements					
Biotin only	−	−	+	−	+/−
Biotin, calcium pantothenate, and nocotinic acid	−	−	−	+	−
3-Ketolactose production	+	+	−	−	−
Growth on 2% NaCl	+	+	−	+	+
Acid produced from:					
Mannitol	+	+	+	+	+
Adonitol	+	+	+	+	+
Erythritol	−	−	+	−	−
Dulcitol	+	+	+	−	−
Melezitose	+	+	−	−	−
Ethanol	+	+	−	−	−/+
Arabitol	+	+	−	−	−
Alkali produced from sodium L-tartate	−	−	+	−	+
Growth at 37℃	+	+	−	+	−

The species *A. vitis* comprises strains previously referred to as *Agrobacterium* biovar 3. Strains are generally isolated from grapevines but have also been reported from chrysanthemums. The host range is not limited to grapevines in most cases but includes a variety of dicotyledonous plants. Strains may be tumorogenic or nontumorigenic, but to date no shizogenic isolates have been described. All strains that have been tested are also capable of causing the

watery dacay on grapevine roots. The type strain is strain K309 (=NCPPB 3554). It requires biotin for growth. The G+C content of the DNA is 59 mol%. Isolated from grapevines in South Australia in 1977, strain NCPPB 3554 is tumorigenic on grapevine shoots, on tomatoes, on carrot disks, and on sunflowers. Galls incited by strain NCPPB 3554 contain octopine, and this strain catabolizes octopine as a sole carbon and nitrogen source.

■ Recipes for selective media.

1) Medium 1A for biovar 1

	per L
L (-) arabitol	3.04 g
NH_4NO_3	0.16 g
KH_2PO_4	0.54 g
K_2HPO_4	1.04 g
Sodium taurocholate	0.29 g
$MgSO_4.7H_2O$	0.25 g
Agar	15.00 g
Crystal violet, 0.1% (w/v) aqueous	2 ml

Autoclave, cool to about 50 ℃, then add filter-sterilized cycloheximide (1.0 ml of 2% solution) and Na_2SeO_3 (6.6 ml of 1% aqueous).

2) Medium 2E for biovar 2

	per L
NH_4NO_3	0.16 g
Erythritol	3.05 g
KH_2PO_4	0.54 g
K_2HPO_4	1.04 g
$MgSO_4 \cdot 7H_2O$	0.25 g
Sodium taurocholate	0.29 g
Yeast extract, 1% (w/v) aqueous	1 ml
Malachite green, 0.1% (w/v) aqueous	5 ml
Agar	15.00 g

Autoclave, cool to 50 ℃, then add filter-sterilized cycloheximide (1.0 ml of 2% solution) and Na_2SeO_3 (6.6 ml of 1% aqueous)

3) Roy-Sasser medium for biovar 3

	per L
$MgSO_4 \cdot 7H_2O$	0.2 g
K_2HPO_4	0.9 g
KH_2PO_4	0.7 g
Adonitol	4.0 g
Yeast extract	0.14 g
NaCl	0.2 g
H_3BO_3	1.0 g
Agar	15.0 g
Chlorothalonil (Bravo 500), 4% (w/v) aqueous	0.5 ml

Adjust to pH 7.2, autoclave, cool to 50 ℃, and add aseptically the following (after dissolving separately in a few ml of distilled water and filter sterilizing):

Triphenyltetrazolium chloride	80 mg
D-cycloserine	20 mg
Trimethoprim (with 1 drop HCl to the distilled water)	20 mg

Colonies of biovar 3 are countable after four days of incubation at 27 ℃. They will have dark red centers with white edges. Comparison to a known isolate plated at the same time is recommended.

3. 식물병원성 Pseudomonads의 재분류

지금까지 *Pseudomonas* species는 주로 형광 물질의 생성과 poly-β-hydroxybutylate inclusion의 생성에 근거하여 분류되어져 왔다. 그러나 최근 수년간 새로운 분자 생물학적 방법과 생리 생화학적 방법에 의거하여 *Pseudomonas* genus가 여러 개의 새로운 genus로 재분류되어야 한다는 많은 보고가 있었다. 1988년에 발표된 Approved list of bacterial names 에는 86종이 genus *Pseudomonas*에 속해있다. 그중 32종이 식물병원 세균이다. 1980년대 말 Palleroni등은 DNA-DNA, DNA-rRNA hybridization 방법을 이용하여 *Pseudomonas*를 5개의 rRNA subgroup으로 나누었다; 1) *Pseudomonas aeruginosa* group, 2) *P. solanacearum*, 3) *P. acidovorous*, 4) *P. diminuta*, 그리고 5) *P. maltophilia*. 이러한 재분류는 많은 다른 방법에 의해서도 확인. 입증되었으며, 그중 cellular fatty acid 분석에 의하면, 각 rRNA subgroup은 각각의 독특한 fatty acid profile을 가지고 있었다. 그 뒤 Stead는 fatty acid profile을 이용하여 *Pseudomonas*를 다시 6개의 subgroup으로 나누었으며, 이 subgroup은 앞에서의 rRNA에 의한 subgroup과 비슷하다. 그러나 최근 개발된 phylogenetic classification을 종합 고찰해 보면 genus *Pseudomonas*는 너무나 다양한 세균의 모임이 되어 새로운 genus로의 재분류가 요구되어져 왔다. 이러한 요구에 의거하여 많은 연구가 행해졌으며, 새로운 genus가 제안됐고 또 받아들여졌다. 이러한 제안은 cellular lipid and fatty acid analyses, DNA-DNA hybridization, DNA-rRNA hybridization, 그리고 1,174 개의 16S rRNA를 coding 하는 DNA의 sequence alignment 등에 의하여 행해졌다. P. De Vos는 *Hydrogenophaga, Acidovorax, Comamonas, Stenotrophomonas, Burkholderia, Brevundimonas*등의 genus를 포함하는 새로운 family Comamonadaceae를 제안하였으며, committee에 의하여 받아들여졌다. 하지만 아직도 많은 *Pseudomonas*에 속해있는 세균이 잘못 분류되었으며, 재분류되어져야 한다.

※ A key for the differentiation of the different groups in the *Comamonadaceae*

I. Not maintainable on nutrient agar
   A. Yellow pigmented, very slow growth in general, and oxidase absent: *Xylophilus*
   B. Unpigmented, oxidase present: [*Aquaspirillum*] species

II. Good growth on nutrient agar
   A. Yellow pigmented and usually capable of chemolithotrophic growth with hydrogen
      1. Grows with L-arabitol, mesaconate, and citrate as sole carbon sources: *Varivorax*
      2. Does not use L-arabitol, mesaconate, and citrate as sole carbon sources: *Hydrogenophaga*
   B. Unpigmented, no chemolithotrophic growth with hydrogen
      1. Bipolar tufts of flagella, does not grow with D-glucose as a sole carbon source: *Comamonas*
      2. One polar flagellum, grows with D-glucose as a sole carbon source: *Acidovorax*

(1) GROUP 4. GRAM-NEGATIVE AEROBIC/MICROAEROPHILIC RODS AND COCCI

○ Genus *Pseudomonas*

Straight or slightly curved rods, but not helical, 0.5-1.0 x 1.5-5.0 μm. Many species accumulate poly-β-hydroxybutylate as carbon reserve material, which appears as sudanophilic inclusions. They do not produce prosthecae and are not surrounded by sheaths. No resting stages are known. Cells stain Gram negative. Motility occurs by one or several polar flagella; they are rarely nonmotile. In some species lateral flagella of shorter wavelength may also be formed. Aerobic, having a strictly respiratory type of metabolism with oxygen as the terminal electron acceptor; in some cases nitrate can be used as an alternate electron acceptor, allowing growth to occur anaerobically. Xanthomonadins are not produced. Most, if not all, species fail to grow under acidic conditions (pH 4.5). Most species do not require organic growth factors. Oxidase positive. Catalase positive and chemoorganotrophic; some species are facultative chemoorganotrophs, able to use H2 or CO as energy sources. Widely distributed in nature. Some species are pathogenic for humans, animals, or plants.

The two main groups of phytopathogenic pseudomonads are 1) the fluorescent group, which produce a fluorescent pigment on KB, and which usually lacks poly-β-hydroxybutylate (PHB) accumulation but does not grow on D-arabinose, or 2) the non-fluorescent group which usually accumulates PHB and uses D-arabinose but not produce a fluorescent pigment. However, some strains, pathovars , and a few species which genotypically belong to the fluorescent group do not produce the fluorescent pigment on KB so that lack of pigment production does not necessarily place an organism in the non-fluorescent group.

Recent reclassification of psedomonads grouped several non-fluorescent phytopathogenic *Pseudomonas* species into several new genus. However, most fluorescent *Pseudomonas* species are still in the same genus.

Psuedomonas avenae → *Acidovorax avenae*
Pseudomonas solanacearum → *Burkholderia solanacearum* → *Ralstonia solanacearum*
Pseudomonas androphogonis → *Burkholderia androphogonis*
Pseudomonas caryophylli → *Burkholderia caryophylli*
Pseudomonas cepacia → *Burkholderia cepacia*
Pseudomonas gladioli → *Burkholderia gladioli*
Pseudomonas plantarii → *Burkholderia plantarii*
Pseudomonas glumae → *Burkholderia glumae*
Xanthomonas ampelina → *Xylophilus ampelinus*

Table Sources and characteristics of additional *Pseudomonas* species [a]

SOURCE	SPECIES	CHARACTERISTICS AND REFERENCES
ISOLATED FROM DISEASED PLANTS AND MUSHROOMS		
Cultivated mushrooms	*P. agarici*	One, rarely two, flagella. Fluorescent pigment produced. Acid is produced from glucose and various other sugars. Oxidase positive. **Causes drippy gill of mushrooms**. One of the main differences with another mushroom pathogen, *P. tolaasii*, is in the utilization of benzoate. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 188.
	P. tolaasii	One to five flagella. Fluorescent pigment produced. Acid is produced from glucose. Gelatin is liquefied. **Isolated from brown spot of cultivated mushrooms**. See *Bergey's Manual of Determinative Bacteriology*, 7th ed., p. 136.
Birds-nest fern	*P. asplenii*	One to three flagella. Fluorescent pigment produced. Temperature range: 1–34°C. Acid is produced from glucose and various other sugars. Gelatin is liquefied. **Isolated from lesions of the bird's nest fern (*Asplenium nidus*)**. See *Bergey's Manual of Determinative Bacteriology*, 7th ed., p. 124.
Pawpaw	*P. caricapapayae*	Three to six flagella. Fluorescent pigment produced. Temperature range: 7–45°C. Acid is produced from glucose and various other sugars. Gelatin is liquefied. **Isolated from water-soaked, angular spots on leaves of pawpaw**. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 188.
Almond tree	*P. amygdali*	One to six flagella. No fluorescent pigment produced. Temperature range: 3–32°C. Acid is produced from glucose and various other sugars. Gelatin is not hydrolyzed. **Produces a hyperplastic bacterial canker in the almond tree (*Prunus dulcis*, fam. *Rosaceae*)**. Not pathogenic for other fruit trees. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 188–189.
Ficus erecta tree	*P. ficuserectae*	Motile by 1–5 flagella. Poly-β-hydroxybutyrate is accumulated. Pigments are not produced. Similar to *P. amygdali* in many properties but differs by forming larger colonies on nutrient agar, utilizing raffinose and glycerol, and failing to hydrolyze Tween 80, to produce H_2S, and to utilize ribose, mannitol, and sorbitol. **Causes dark brown, water-soaked spots on the leaves and stems of *Ficus erecta*** Thumb., resulting either in defoliation or shoot blight on severely infected plants. For other characteristics see Goto, Int. J. Syst. Bacteriol. *33*: 546–550, 1983.
Sorghum, corn, clover, and velvet bean	*P. andropogonis*	One, rarely two, flagella. Sheathed flagella have been reported in some strains. No fluorescent pigment produced. Poly-β-hydroxybutyrate is accumulated. Most strains are oxidase negative. Glucose and various other sugars are utilized. Gelatin is not hydrolyzed. The species may be divided into two specialized pathovars, namely, pv. *andropogonis*, the agent of a **stripe disease of sorghum**, and pv. *stizolobii*, which has been described as the cause of **leaf spot of velvet bean** (*Stizolobium deeringianum*). See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 189.
Oats (*Avena sativa*) and foxtail (*Chaetochloa lutescens*)	*P. avenae*	No fluorescent pigment produced. Poly-β-hydroxybutyrate is probably accumulated. Oxidase negative. Acid is produced from glucose and various other sugars. Gelatin liquefaction is variable. **Pathogenic for oats (*Avena sativa*) and foxtail (*Chaetochloa lutescens*)**. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 189.

Table *(continued)*

SOURCE	SPECIES	CHARACTERISTICS AND REFERENCES
Orchids	*P. cattleyae*	One or two bipolar flagella. No fluorescent pigment produced. Acid is produced from glucose and various other sugars. Gelatin is not liquefied. **Pathogenic for *Cattleya* sp. and *Phalaenopsis* sp.** (fam. *Orchidaceae*). See *Bergey's Manual of Determinative Bacteriology*, 7th ed., p. 148.
Cissus plants	*P. cissicola*	Non-motile immediately after isolation, but motile clones appear after subculturing, the cells of which have a polar flagellum. Poly-β-hydroxybutrate is accumulated. No fluorescent pigment is produced. No growth below 5°C or above 37°C. Acid is produced from glucose and various other sugars by most isolates. Gelatin is liquefied. Starch is hydrolyzed. Arginine dihydrolase is negative. **Pathogenic for *Cissus japonica*** (fam. *Vitaceae*). See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 189.
Tomato plants	*P. corrugata*	Multitrichous polar flagella. Poly-β-hydroxybutyrate is accumulated. No fluorescent pigment is produced. Yellow-green diffusible, non-fluorescent pigment is produced. Colonies are wrinkled, yellowish, sometimes with green center. Growth occurs at 37°C but not at 41°C. Gelatin is hydrolyzed. Among the characters that differentiate this species from *P. cepacia* and *P. gladioli* are the absence of pectate hydrolysis and rot of onion slices, and the lack of utilization of D-arabinose, cellobiose, adipate, *meso*-tartrate and citraconate. **Isolated from tomato pith necrosis.** See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 189.
Rice	*P. glumae*	Two to four flagella. Fluorescent pigment produced on potato agar. Temperature range: 11–40°C. Acid is produced from glucose and various other sugars. **Pathogenic for rice plants** (*Oryza sativa* fam. *Gramineae*). See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 189–190.
	P. fuscovaginae	Motile by 1–4 polar flagella. Produce a green, fluorescent, diffusible pigment. Oxidase positive. No growth at 37°C. Unable to denitrify. Acid is produced from glucose and various other carbohydrates. Arginine dihydrolase positive. **Distinguished from other arginine dihydrolase positive fluorescent pseudomonads by its ability to produce a hypersensitivity reaction in tobacco plants and its inability to utilize 2-ketogluconate or inositol. Pathogenic for *Oryza sativa, Hordeum vulgare, Triticum aestivum, Avena sativa, Zea mays, Lolium perenne, Bromus marginatus, Phleum pratense*, and *Phalaris arundinacea*.** For other characteristics see Miyajima et al., Int. J. Syst. Bacteriol. *33*: 656-657, 1983.
	P. plantarii	Motile by 1–3 polar flagella. Poly-β-hydroxybutyrate is accumulated. Colonies have a slight yellow tint and weakly produce a diffusible, reddish brown pigment under certain conditions. Fluorescent pigments not produced. Oxidase positive. Organic growth factors not required. Able to denitrify. Arginine dihydrolase negative. Gelatin is liquefied. Temperature range: 4–10°C to 38°C. **Tobacco hypersensitivity reaction negative.** Acid is produced from glucose and various other carbohydrates. **Causes seedling blight of rice.** For other characteristics see Azegami et al., Int. J. Syst. Bacteriol. *37*: 144-152, 1987.
Sugarcane	*P. rubrilineans*	Motile by a single flagellum. Poly-β-hydroxybutyrate is accumulated. No pigments are produced. Oxidase positive. Gelatin liquefaction is weak. Capable of growth at 40°C. Acid is produced from glucose and various other sugars. **The agent of red stripe of sugarcane.** See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 190.

Table **Sources and characteristics of additional *Pseudomonas* species** [a]

SOURCE	SPECIES	CHARACTERISTICS AND REFERENCES
	P. rubrisubalbicans	Slightly curved rods motile by several polar flagella. Poly-β-hydroxybutyrate is accumulated. No pigments are produced. Oxidase positive. Gelatin is not hydrolyzed. Capable of growth at 40°C. Acid is produced from glucose and various other sugars. **The agent of mottled stripe of sugarcane**. See *Bergey's Manual of Systematic Bacteriology*, Vol. 1, p. 190.
Carnation	*P. woodsii*	Motile by a single polar flagellum. Gelatin is not liquefied. Acid is produced from glucose and various other sugars. Isolated from water-soaked lesions on carnation leaves. **Pathogenic for carnation** (*Dianthus caryophyllus*, fam. *Caryophyllaceae*). See *Bergey's Manual of Determinative Bacteriology*, 7th ed., p. 150-151.

Table The fluorescent pigment producing species

Test	*Pseudomonas*					
	marginalis	*tolaasii*	*agarici*	*cichorii*	*viridiflava*	*syringae(GI)*
Levan	+	−	−	−	−	V
Oxidase	+	+	+	+	+	−
Arginine dihydrolase	+	−	−	−	−	−
Nitrate to N_2	−	−	−	−	−	−
Growth at 41 ℃	−	−	−	−	−	−
Potato rot	+	−	−	−	+	−
Used for growth:						
Mannitol	+	+	+	+	+	V
Geraniol	−	−	−	−	−	−
Benzoate	−	−	+	−	−	−
Cellobiose	−	−	−	−	−	−
Sorbitol	+	+	−	−	+	V
Trehalose	+	V	−	−	−	−
Sucrose	+	−	−	−	−	V
m-Tartrate	V	+	−	+	+	V
D-Tartrate	V	−	−	−	+	V
D-Arabinose	−	−	−	−	−	−
L-Rhamnos	V	ND	−	−	−	−
D-Aspartate	−	ND	ND	+	−	−

○ Geus *Acidovorax*

Straight to slightly curved rods, 0.2-0.7 x 1.0-5.0 μm, occurring singly or in short chains and motile by means of a single polar flagellum. Cells stain Gram negative. They are oxidase positive. Urease activity varies among strains. Some strains grow on Christensen urea agar but lack urease according to API 20NE tests. No pigment is produced on nutrient agar. Aerobic and chemoorganotrophic. *Acidovorax facilis* and several *Acidovorax delafieldii* strains are capable of lithoautotrophic growth by using hydrogen as an energy source. Oxidative carbohydrate metabolism occurs with oxygen as the terminal electron acceptor; alternatively, some strains of *Acidovorax delafieldii* and *Acidovorax temperans* are capable of heterotrophic denitrification of nitrate. Good growth is obtained on media containing organic acids, amino acids, or peptone, but only a limited number of sugars are used for growth. Two hydroxylated fatty acids, 3-hydroxyoctanoic acid (3-OH-8:0) and 3-hydroxydecanoic acid (3-OH-10:0), are always present, 2-hydroxylated fatty acids are absent, and a cyclopropane-substituted fatty acid (17:cyc) is present in most of the strains. The mean G+C values of the DNAs are 62-66 mol%.

Table. Differential characteristics of the species of the genus *Acidovorax*

Characteristics	*A. delafieldii*	*A. facilis*	*A. temperans*
Growth on :			
L-arabinose, D-galactose, D-ribose, D-mannose	+	+	−
Adipate	+	−	+
2-Ketogluconate, 2-ketoglutarate, citraconate	+	−	d
NO_2 - reduction	d	−	+
Gelatinase	d	+	−
Autotrophic growth with hydrogen	d	+	−

○ Genus *Burkholderia*

The cells of species of this genus are Gram-negative, nonfermentative, straight rods, that have a single polar flagellum or tuft of polar flagella when motile. Catalase is produced, oxidase activity is variable by species. Cellular lipids are characterized by the presence of phosphatidylglycerol possessing hydroxy fatty acid at the 2-position of glycerol. Fatty acid compositions of cellular lipids are characterized by the presence of 2OH acids of C16:0, 16:1, 18:1, 19CPA and 3OH acids of C14:0 and 16:0 and, characteristically absent are 2OH-C12:0, 3OH-C10:0 and 3OH-C12:0. Species are pathogenic to either human or plant. G+C content of DNA was 64-68 mol%. (E. Yabuuchi et al., 1992. Proposal of *Burkholderia* gen. nov. and Transfer of Seven Species of the Genus *Pseudomonas* Homology Group II to the new Genus, with the Type Species *Burkholderia cepacia* (Palleroni and Holmes 1981) comb. nov. Microbiol. Immunol. Vol.36:1251-1275)

○ Genus *Ralstonia*

The cells of species of this genus are Gram-negative rod shaped, motile with single polar or peritrichous flagella or non-motile without flagella. Oxidase activity is variable by species. The absence of assimilation of galactose, mannitol, mannose, and sorbitol by the type strain of three *Ralstonia* species is a differential phenotypic marker to distinguish strains of *Ralstonia* sp. from those of *Burkholderia* species. Cellular lipids of the type strains of three *Ralstonia* species contain phosphoatidylethanolamine possessing 2-hydroxy fatty acid at the sn2 position of the glycerol moiety, similar to those of *Burkholderia* species. The type strains of three *Ralstonia* species failed to demonstrrate two kinds of ornithine-lipids on two dimensional thin layer chromatography and to reveal C19 cyclopropanoic acid in cellular fatty acids. The presence of these cellular lipids and fatty acid components were clearly demonstrated in *Burkholderia cepacia* and the other five *Burkholderia* species.

○ Genus *Xylophilus*

Straight to slightly curved rods, 0.4-0.8 x 0.6-3.3 μm. Filamentous cells (length, 30 μm or more) may occur in older cultures. Cells occur singly, in pairs, or in short chains. Motility occurs by a single polar flagellum. Cells stain gram negative. Oxidase negative, catalase positive. Aerobic, chemoorganotrophic. Oxidative carbohydrate metabolism. Even at the optimal temperature of 24 ℃, growth is generally very slow and poor. Growth occurs on L-glutamine but no on calcium lactate (in contrast to *Xanthomonas* strains). Plant pathogens, causing bacterial necrosis and canker.

Table Phenotypic features of 6 type strains of 3 *Ralstonia* sp., 2 *Burkholderia* sp. and *A. faecalis*

	R. pickettii EY 3254^T	*R. solanac.* EY 2181^T	*R. eutropha* EY 3798^T	*B. andropogonis* EY 3792^T	*B. cepacia* EY 645^T	*A. faecalis* EY 1056^T
Gram-negative rod-shaped	+	+	+	+	+	+
Motility	+	−	+	+	+	+
Flagellation	Polar mono	None	Peri	Polar mono	Polar tuft	Poorly peri
Catalase	+	+	+	+	+	+
Oxidase, Kovacs	+	+	+	+	−	+
Growth at 41 C	+	−	+	−	+	
37 C	+	−	+	+	+	+
Growth: MacConkey agar	−	−	+	+	+	+
NaCl 0%	+	+	+	+	+	+
NaCl 3%	−	−	+	+	+	
NaCl 5%	−	−	−	−	+	
Sheep blood agar, green	−	−	+	−	+	+
Fermentation of glucose, KI agar	−	−	−	−	−	−
Blackening of KI butt	−	−	−	−	−	−
Citrate, Simmons	−	+	+	+	+	+
Citrate, Christensen	−	+	+	+	+	+
Nitrate to gas	+	−	−	−	−	−
NO_3 to NO_2	+	+	+	−	−	−
Zn test on negative nitrite test	NT	NT	NT	+	+	+
Malonate	+	+	+	+	+	+
Hydrolysis of esculin	−	−	−	−	+	−
Liquefaction of gelatin	+	+	−	−	+	−
Hydrolysis of Tween 80	−	+	+	−	+	−
DNase	−	−	−	−	−	−
Phenylalanine deaminase	+	−	−	+	+	−
Urease	+	+	−	+	−	−
Arginine dihydrolase, Moeller	−	−	+	−	−	−
Lysine decarboxylase, Moeller	−	−	−	−	+	−
Ornithine decarboxylase, Moeller	−	−	−	−	+	−
Carlquist ninhydrine test	−	−	−	+	+	−
Acylamidase	−	−	−	−	+	−
Oxidative acid from: Adonitol	+	−	−	+	+	−
D- & L-Arabinose	−	−	−	+	+	−
Cellobiose	+	−	−	−	+	−
Dulcitol	−	−	−	−	+	−
Ethanol, trehalose	−	+	−	−	+	−
Galactose	+	−	−	+	+	−
Glucose	+	−	+	+	+	−
Fructose, glycerol, D-xylose	+	+	+	+	+	−
Inositol, sorbitol, mannitol	−	−	−	+	+	−
Inulin	−	−	+	+	−	−
Lactose	+	−	+	+	+	−
Maltose	+	+	−	−	+	−
Mannitol	−	−	−	+	+	−
Mannose	+	+	−	+	+	−
Melezitose, melibiose	−	−	−	−	−	−
Raffinose, rhamnose	−	−	−	−	−	−
D-Ribose	+	−	+	+	+	−
Salicin	−	−	−	−	−	−
Sucrose	−	+	+	−	+	−
OF base alkaline	+	+	+	+	+	+
Major respiratory quinone	Q8	Q8	Q8	Q8	Q8	Q8
Mol% G+C of DNA	64	66.6	66.5	60.1	66.6	57.5

Table Plant pathogenic genera and species

Previous Genus	Present Genus	Species/subspecies [number of pathovars]
Corynebacterium	*Arthrobacter*	*ilicis*
	Rathayibacter	*iranicus*
		rathayi
		tritici
	Clavibacter	*michiganensis* subsp. *michganensis*
		michiganensis subsp. *insidiosus*
		michiganensis subsp. *nebraskensis*
		michiganensis subsp. *sepedonicus*
		michiganensis subsp. *tessellarius*
		xyli subsp. *xyli*
		xyli subsp. *cynodontis*
	Curtobacterium	*flaccumfaciens* pv. *betae*
		flaccumfaciens pv. *poinsettiae*
		flaccumfaciens pv. *flaccumfaciens*
		flaccumfaciens pv. *oortii*
	Rhodococcus	*facians*
Pseudomonas	*Acidovorax*	*avenae* subsp. *avenae*
		avenae subsp. *cattleyae*
		avenae subsp. *citrulli*
		konjaci
	Burkholderia	*andropogonis*
		caryophylli
		cepacia
		gladioli pv. *alliicola*
		pv. *gladioli*
		plantari
		glumae
	Ralstonia	*solanacearum*
	Xylophilus	*ampelinus* (actually from *Xanthomonas ampelina*)

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