

# Diseases associated with pathovars of the *Xanthomonas campestris* group in Puerto Rico<sup>1</sup>

Amelia Cortés-Monllor<sup>2</sup>

## ABSTRACT

A revision of the pathovars of the *Xanthomonas campestris* group affecting cultivars in Puerto Rico is presented. Some have not been previously reported. The pathogen has been isolated from beans, cabbage, cotton, maize, pepper, pigeon pea, soybean, sugarcane, tanager, tomato and turnip, and from ornamentals *Anthurium*, *Begonia*, *Dieffenbachia* and *Syngonium*. These have been obtained from growing areas with different rainfall and temperatures.

## RESUMEN

Enfermedades asociadas con patovares del grupo *Xanthomonas campestris* en Puerto Rico

Se presenta una revisión de cultivares afectados por patovares de *Xanthomonas campestris*. Algunos patovares se informan por vez primera. El patógeno se ha aislado de habichuelas, repollo, algodón, maíz, pimiento, gandul, soya, caña de azúcar, yautía, tomate y nabo. También de las ornamentales anturio, *Begonia*, *Dieffenbachia* y *Syngonium*. Estos cultivares se obtuvieron de zonas de Puerto Rico con varios factores climáticos.

## INTRODUCTION

*Xanthomonas* species are always associated with plants. These bacteria cause diseases such as leaf blight, black rot, gummosis and wilt. Four species: *Xanthomonas albilineans*, *X. ampelina*, *X. axonopodis*, and *X. fragariae*, have been reported attacking members of only one plant genus. The fifth species, *X. campestris*, includes over 120 pathovars which can be found in a number of different host plants and are differentiated only by host reaction (14, 17).

In Puerto Rico 14 pathovars of *X. campestris* have been identified (table 1). In 1915 Stevenson (1) reported *Bacterium phaseoli* Erw. F. Smith affecting *Phaseolus vulgaris* in Puerto Rico. In 1917, turnip (22) and cabbage (24) were reported to be affected by black rot caused by *Bacterium campestris* Pammel Erw. F. Smith.

From 1919 to 1933 (2), the gumming disease of sugarcane spread with remarkable rapidity throughout the island. Matz (2) discovered the gummy exudation in the cut ends of the cane stalk. He isolated and

<sup>1</sup>Manuscript submitted to Editorial Board 16 August 1991.

<sup>2</sup>Associate Bacteriologist, Department of Crop Protection.

studied the organism *Bacterium vasculorum* (Cobb) Dye, which was found affecting most of the varieties planted. Red stripe (*X. rubrilineans*, Lee et al.) and mottle stripe (*X. rubrisubalbicans*, Christopher and Edgerton) were also found affecting sugarcane but were not considered as a severe disease (2). Among other crops affected by *X. campestris* pathovars were corn (*X. stewartii*, Erw. F. Smith, Dowson), pepper (*X. vesicatoria*, Doidge), cotton (*X. malvacearum*, Erw. F. Smith) and *Dieffenbachiae* spp. (*X. dieffenbachiae*, McCull and Pirone) (2,23).

Early reports were based on observation of symptoms and not on the behavior of the pathogen. Stevenson (23) states that Pérez and Cortés-Monllor (18) in 1960 reported for the first time on the characterization of *X. vesicatoria* affecting tomatoes in Puerto Rico. Since then, many bacterial diseases caused by other *X. campestris* pathovars have been isolated from different sources and locations. Information has been obtained concerning the identity of many of these pathovars and the symptoms and diseases they cause.

This compendium presents information that would be valuable to scientists and students dealing with plant diseases caused by *X. campestris* pathovars. Included are some pathovars not previously reported.

#### HOSTS AND SYMPTOMS

Disease symptoms due to the *X. campestris* group are observed mostly on leaves, fruits and other plant parts. Usually lesions are at first circular minute yellow dots with a necrotic center. They may enlarge and coalesce forming irregular areas surrounded by a yellow border and may spread toward the midrib and eventually down to the stem. Water-soaked lesions may also soften the tissue; afterwards the plant wilts and, finally, the entire leaf or plant collapses. In some cases exudates emerging from the lesion can be observed early in the morning. Systemic infection can also occur, resulting in symptoms of marginal leaf necrosis, necrotic spots, chlorosis of older leaves, wilting, deformation and stunting. Some species develop very specific symptoms. Blackening of the leaf veins and vascular bundles is often observed on plants belonging to the Brassica group. This becomes clearly evident when the petioles and larger veins are cut transversely. In tomato and pepper fruits (Solanaceae) sunken necrotic lesions with a central pustule are usually the most noticeable symptom.

Bacterial blight of anthurium (*Anthurium andreanum* Lind.) caused by *X.c. dieffenbachiae*, McCull and Pirone was first reported in Puerto Rico by Sánchez (21) in 1986. Primary symptoms consist of irregular yellow-brown spots on young and adult leaves eventually turning dark brown. Exudate might be observed and some defoliation can occur. On older leaves the most common symptom observed is large necrotic areas

surrounded by a yellow border. Usually these are toward the tip and on the margins of the leaf (plate 1). Exudate might be observed underneath the leaf early in the morning. Systemic infection causes yellowing of the lower leaves followed by necrosis, wilting and death. Drops of bacterial exudate will often appear on the cut surfaces of the stem and short black threads may be seen on the vascular tissue.

Common blight of beans (*Phaseolus vulgaris* L.) is caused by *X.c. phaseoli* Erw. F. Smith. In 1915, Stevenson (1) in Puerto Rico reported it as one of the most serious diseases of beans. Leaf-hoppers and other insects were the chief factor of the spreading of the disease. Bacterial lesions are initially small necrotic water-soaked leaf spots, irregular in shape, surrounded by a bright yellow margin. Some spots may coalesce to form large necrotic areas which dry out the entire leaf; defoliation often follows. Dark green vertical spots on the stem gradually change to brownish. The lesions on pods are necrotic, sunken and watery. Seeds may become infected. Symptoms can be confused with those of the halo-blight disease caused by *Pseudomonas phaseolicola*.

Bacterial leaf spot of begonia (*Begonia decandra* Pavon) is caused by *X.c. begonia* (Takimoto) Dowson. In 1971, Cortés-Monllor isolated the organism from diseased plants in the Naranjito nursery (table 1; fig. 4). Initial symptoms are small water-soaked or dry lesions on the margin of the leaves. They enlarge and turn necrotic extending toward the center area. On systemic infection the plant wilts and collapses.

Black rot of cabbage (*Brassica oleracea* L.) is caused by *X.c. campestris* (Pammel) Dowson. In 1917, Thomas (24) reported cabbage plants affected by a severe disease caused by *Pseudomonas campestris*. Later Cook (2) in 1934, reported the disease was seed-transmitted and caused by *Bacterium campestris* (Pammel, EFS). The most noticeable symptom is the yellowish dry area along the margin of the leaves with blackening veins sometimes in a network pattern (plate 1). A cross section of the base of the plant shows black dots extending as black threads within the affected area (plate 1). Exudate may appear in a short time. Seed transmission will result from systemic invasion. Diseased seedlings and young plants collapse in the early stages. Sometimes bacterial soft rot caused by *Erwinia carotovora* will develop.

Bacterial blight of cotton (*Gossypium hirsutum* L.) is caused by *X.c. malvacearum* Erw. F. Smith. Symptoms described by Cook in 1934-35 (2) were angular leaf spots, black-arm of stems and black lesions and gummosis of bracts and bolls of *Gossypium*.

Leaf spot of *Dieffenbachia* (*D. pictae* Schott). Reported in 1939 (23) affecting *Dieffenbachiae* spp. Leaves present small yellow-brown spots followed by yellowing, wilting and necrosis of the entire area. The causal agent is *X.c. dieffenbachiae*, McCull et Pirone.

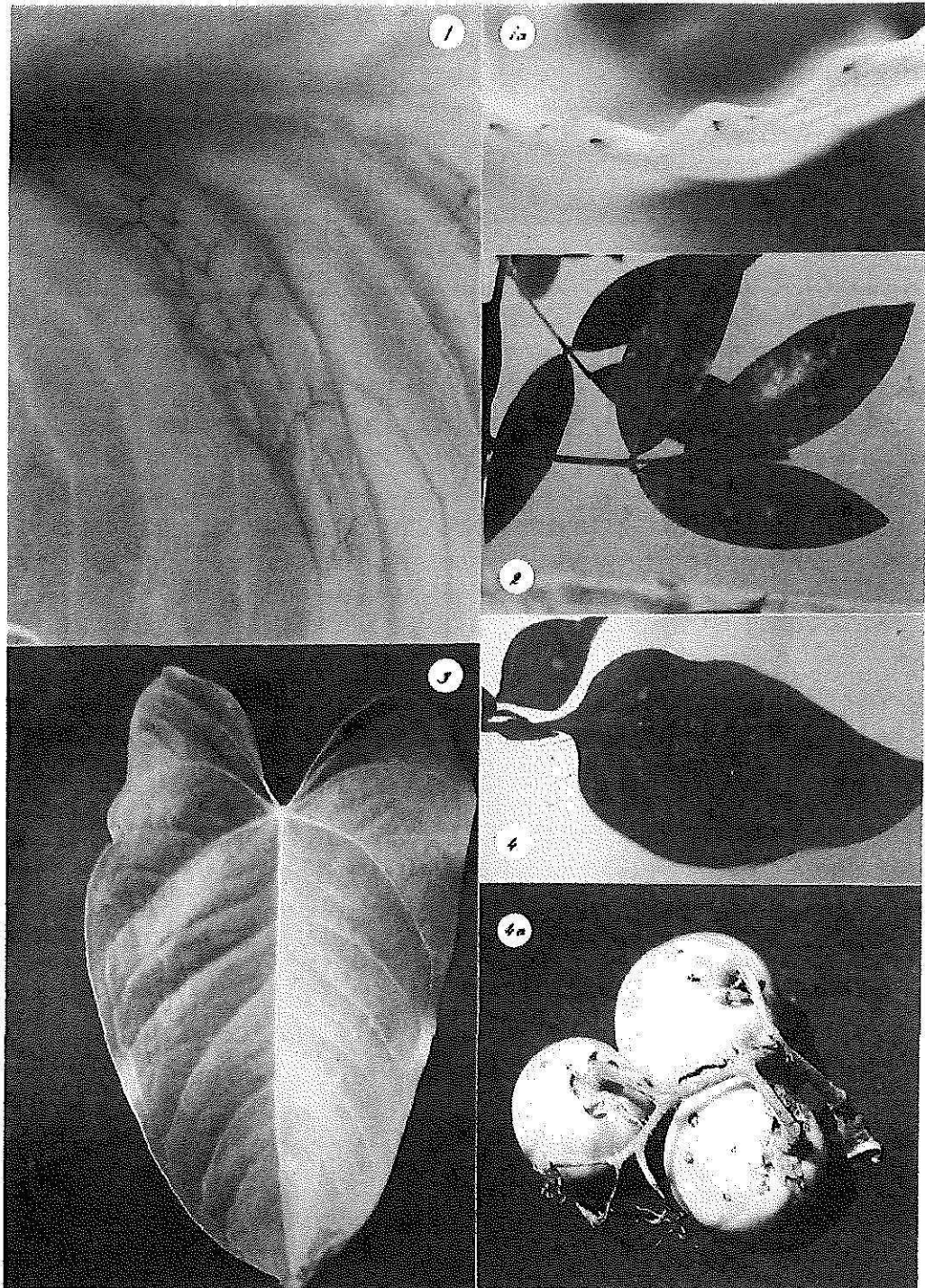


Plate 1

1.—Black rot (black veins) of cabbage caused by *X.c. campestris*.

1a.—Black dots on central vein of cabbage.

2.—Pigeon pea leaf affected by *X.c. cajani*.



Bacterial wilt of maize (*Zea mays* L.) was reported in Puerto Rico (2) caused by *Aplanobacter stewartii* (Erw. F. Smith), McCulloch, severely attacking plantings of hybrid corn growing in low ground. Elliott (15) described this organism isolated in Puerto Rico as *Bacterium stewartii* (Erw. F. Smith), Dowson. Dowson (13) mentioned that it caused considerable damage and losses in some areas of Puerto Rico and other maize-growing areas in the United States. It causes a vascular wilt disease which exudates a bright yellow slime. Affected plants show pale yellow stripes on the leaves; they may show dwarfing and wilting. Plants may eventually die. At present this organism is classified as *Erwinia stewartii* (Erw. F. Smith), Dye (6).

Leaf blight of Nephthytis (*Syngonium podophyllum* Schott), recently reported in Puerto Rico by Cortés-Monllor (10), is caused by *X.c. syngonii* Dickey and Zunoff. The blight is characterized by dry irregular brownish lesions surrounded by a yellow rim. In some cases water-soaked spots can develop in the interveinal areas extending toward the mid-rib down to the petiole (plate 2). Eventually the leaf becomes pale yellow and dies.

Bacterial scab (bacterial pustule) of pepper (*Capsicum annumm* L.) and tomato (*Lycopersicon esculentum* Mill.) caused by *X.c. vesicatoria*, Doidge (plate 1) has been frequently observed in Puerto Rico (2,18,26). The initial symptoms are water-soaked dark-green dots on the underside of the leaves. Gradually the dots enlarge and turn brownish, irregular and slightly raised. In many cases the young leaves will become chlorotic and slightly deformed. On systemic infection the stem will show longitudinal brownish dry lesions, usually crateriformed. The disease is most noticeable on green fruits where lesions are at first small and water-soaked. Gradually the lesions become slightly raised turning light brown and irregular in shape. Eventually the spots enlarge leaving a concave or sunken appearance with a central pustule or blister. Usually they appear to be dry or scabby. On green peppers the spots are surrounded by a pale yellow halo.

Leaf spot of pigeon pea (*Cajanus cajan* (L) Millsp.), reported in Puerto Rico in 1987 (11), is caused by *X.c. cajani*, Kulkarni et al. The symptoms on the lower leaves present minute water-soaked lesions with a necrotic dot in the center, surrounded by a yellow halo. Some of the spots coalesce forming large necrotic areas. Occasionally stem and petioles present longitudinal necrosis (plate 1).



- 3.—Anthurium leaf affected by *X.c. dieffenbachiae*.
- 4.—Lesions incited on pepper leaf by *X.c. vesicatoria*.
- 4a.—Pustules incited by *X.c. vesicatoria* on tomato fruit.

Bacterial pustule of soybean [*Glycine max* (L) Merr.] is caused by *X.c. glycines* (Nakamo). The organism was previously classified as *X. phaseoli* var. *sojensis* (Hedges) (6). In Puerto Rico, Leypon (16) isolated the pathogen from various soybean farms affected with the disease. The leaf spots are usually small and necrotic with a very narrow yellow halo. A darker dot can be observed in the center area. Sometimes the tissue becomes cracked. Lesions become irregular but remain small. The symptoms can be mistaken as those of bacterial blight caused by *Pseudomonas syringae* pv. *glycinea* (Coerper) (13).

Gummosis or gumming disease of sugarcane (*Saccharum officinarum* L.) caused by *X.c. vasculorum* (Cobb) Dye. The disease was reported in Puerto Rico in 1919 but had been present for many years before (2,23). The leaf presents vertical yellowish stripe, mostly toward the margin and to the tip of the blade. Reddish-brown dots and blotches may be observed in the stripe, with a necrotic appearance. Usually only the leaves are attacked, but when the vascular tissue is invaded the characteristic gumming or yellow bacterial slime at the terminal bud is observed. Dwarfing and decay may also be present.

Red stripe of sugarcane (*S. officinarum* L.), reported in Puerto Rico in 1929 by Cook (2), is caused by *X.c. rubrilineans*, Lee et al. Elliott (15) includes Puerto Rico in her geographical distribution and describes the symptoms as consisting of long narrow dark red longitudinal streaks on the leaves. Initially the streaks are watery, dark green, and spread up and down the leaf, gradually turning bright red. Sometimes they coalesce to form broad bands. Infection of young central shoots results in top rot. At present the causal agent is identified as *Pseudomonas rubrilineans* (Lee et al.) Stapp (3).

Mottle stripe of sugarcane (*S. officinarum* L.) is caused by *X.c. rubrisubalbicans*, Christopher and Edgerton. It was reported in Puerto Rico in 1932 (2) as caused by *Phytomonas rubrisubalbicans*, with symptoms similar to Red stripe. Elliott (15) describes it as a disease of the leaf blade in which the stripes are predominately red with white areas or white margins. The stripes are parallel to the leaf veins. Coalescing stripes form mottled red and white bands. No exudate has been observed on the leaves. It is classified as *Pseudomonas rubrisubalbicans* (Christopher and Edgerton) Krasilnikov (4).

Bacterial spot of tanager, *Xanthosoma sagittifolium* (L.) Schott, reported in Puerto Rico in 1986 (9), is caused by *X.c. aracearum* (Berniac) Dye. The leaf spots are abundant small water-soaked lesions with a necrotic center surrounded by a bright yellow halo. The lesions coalesce forming large necrotic areas usually toward the mid-rib. Gradually the leaf becomes chlorotic and dry (plate 2).

Bacterial necrotic margin of tanager, *X. sagittifolium* (L.) Schott, is caused by *X.c. campestris* (Pammel) Dowson. In 1988, Cortés-Monllor

isolated the organism from marginal necrosis of tanager leaves obtained from a plantation in Isabela (table 1, plate 4). Initially the leaf edge dries suggesting drought or burn effect. A chlorotic area is observed between the necrotic and healthy tissue. Gradually the infection spreads from the leaf margin toward the interior of the blade. Pohronezny et al. (19) reported that these tanager diseases (bacterial spot and bacterial necrotic margin) were caused by *X.c. dieffenbachiae*.

Black rot of turnip (*Brassica rapa* L.), mentioned by Stevenson (22) in 1906 as caused by *Pseudomonas campestris*, is caused by *X.c. campestris* (Pammel) Dowson. Dowson (13) and Elliott (15) describe the disease as a "dry rot of fleshy root" followed by a soft rot disease, blackening of leaf veins, wilting and stunting of the plant.

Table 1 presents a list of the *X.c.* pathovars recorded in Puerto Rico, their source, name of disease, and the most evident symptoms.

#### CULTURAL STUDIES

Isolation procedures, methods and media used in the characterization of some of the organisms have been stated before (10). Included is MacConkey agar (Mc), which is a differential plating medium where Gram positive bacteria (e.g., yellow pigmented *Corynebacterium*) are inhibited (12). After pure bacterial cultures were obtained, morphological, cultural and biochemical characters were studied.

#### IDENTIFICATION

*Xanthomonas campestris* pathovars are slow growers (17). They will produce visible colonies after incubating for 24 hr at 28°C.

On tryptone glucose agar (TGA) colonies are circular, smooth, raised or dome-like, shiny and transparent. The colonies are of a buff tinge changing to pale yellow and eventually becoming butter-yellow. They may become mucoid, like a gum mass or butyrous. The yellow pigment is called xanthomonadins; the extracellular slime is known as xantham gum, which is non-soluble in water (14, 17). Some strains will produce diffusible brownish pigmentation.

On McConkey agar (Mc), bacterial growth appears in 36 to 38 h. Grayish pin-point colonies change to buff color. By the 4th day the colonies will be yellowish brown. Some strains will develop buff and buttery, slimy or butyrous colonies.

On tryptone glucose broth (TGB), the bacterial growth produces a slight turbidity of homogenous appearance. Eventually the broth becomes more cloudy and a faint pellicle and ring is formed on top of the surface. In many cases a light yellow sediment is formed.

The biochemical tests include a wide range of reactions, many of which are essential for identification of the organism. Most of the *X.*

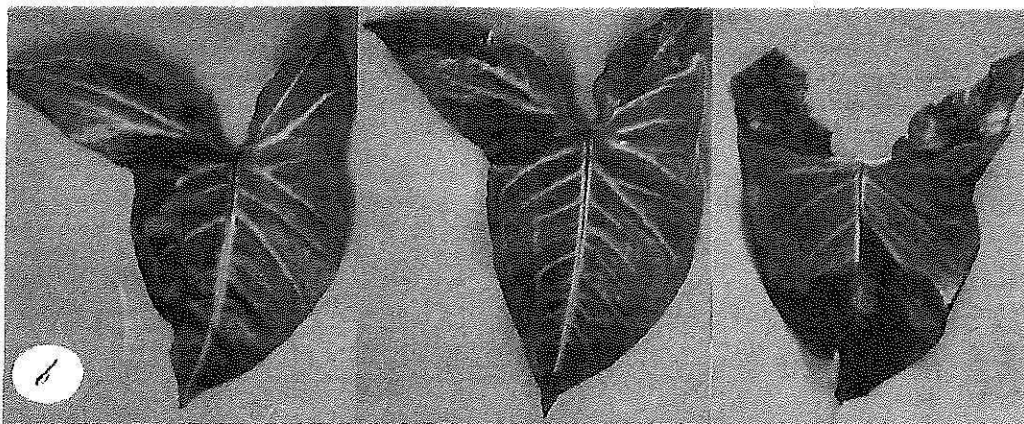
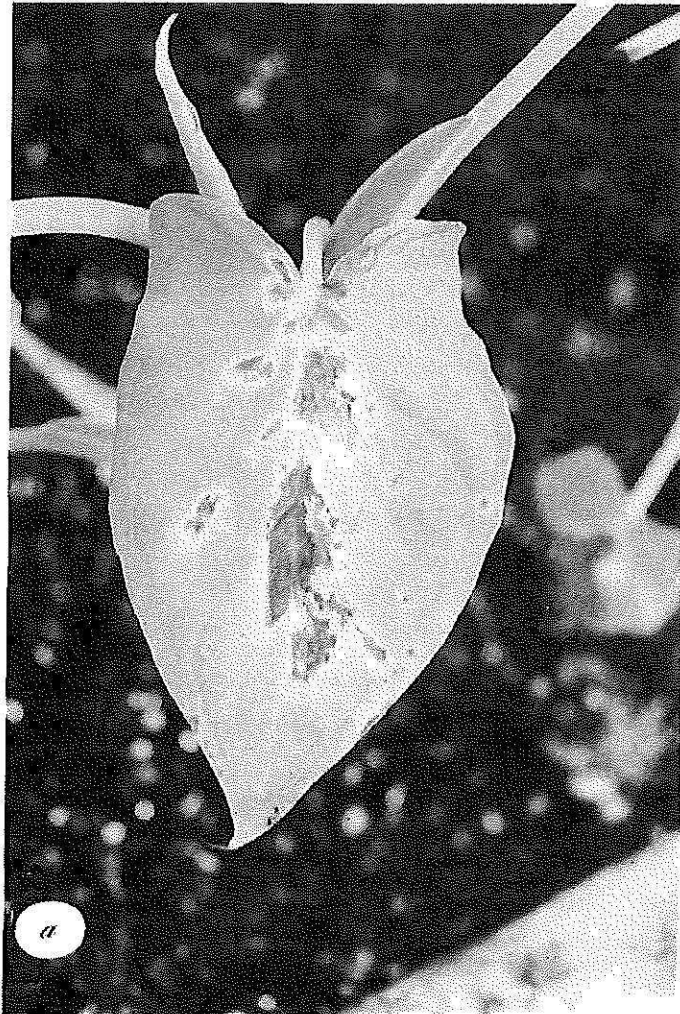
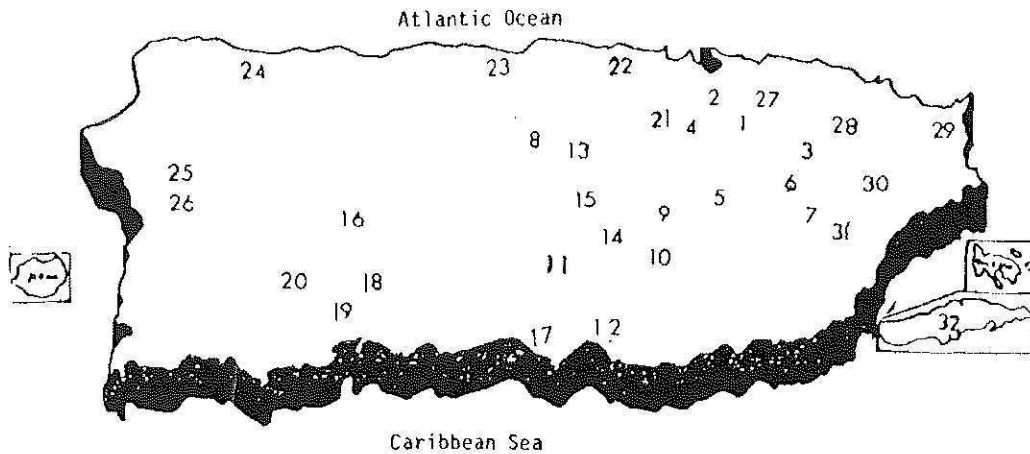


Plate 2

a.—Tanier leaf affected by *X.c. araccarum*.

b.—Advanced symptoms of leaf blight of *Syngonium podophyllum* cv. White Butterfly caused by *X.c. syngonii*.





Locations

- |                  |                  |
|------------------|------------------|
| 1. Trujillo Alto | 17. Santa Isabel |
| 2. Río Piedras   | 18. Peñuelas     |
| 3. Canóvanas     | 19. Guayanilla   |
| 4. Guaynabo      | 20. Yauco        |
| 5. Caguas        | 21. Bayamón      |
| 6. Juncos        | 22. Toa Baja     |
| 7. Las Piedras   | 23. Vega Baja    |
| 8. Morovis       | 24. Isabela      |
| 9. Cidra         | 25. Añasco       |
| 10. Cayey        | 26. Mayagüez     |
| 11. Coamo        | 27. Carolina     |
| 12. Salinas      | 28. Río Grande   |
| 13. Corozal      | 29. Fajardo      |
| 14. Aibonito     | 30. Naguabo      |
| 15. Barranquitas | 31. Humacao      |
| 16. Adjuntas     | 32. Vieques      |

PLATE 3.—Map of Puerto Rico indicating sugarcane areas where gumming disease caused by *X.c. vasculorum* was detected in 1920.

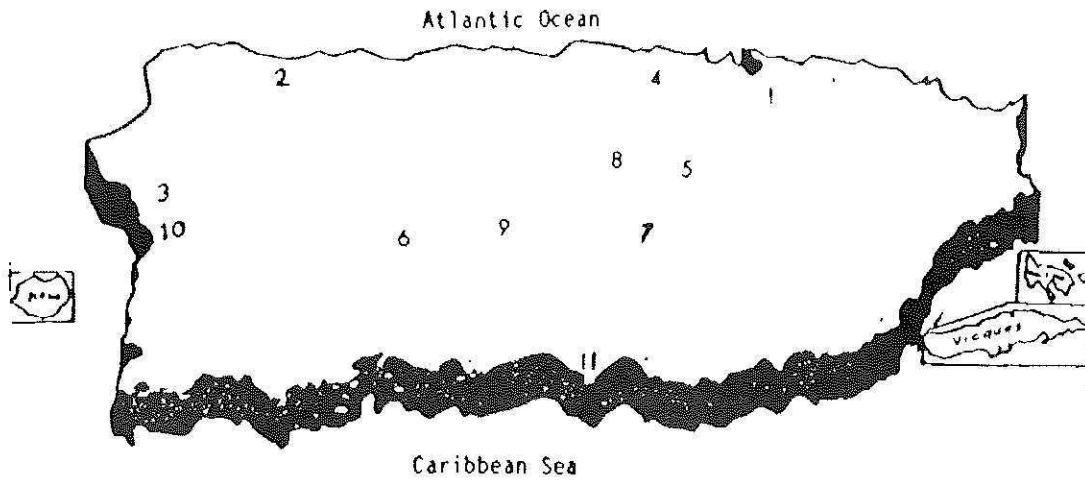
*campestris* pathovars liquefy gelatin, hydrolyze starch and esculin, use citrate and malonate as carbon source, liberate  $H_2S$  from cysteine and peptone and ammonia from peptone. Action in litmus milk varies among the *X. campestris* pathovars. In most cases the initial action occurs when the organism attacks the lacto-albumen to yield ammonia, changing the medium to blue (alkaline). Eventually there is peptonization due to the breakdown or digestion of casein, and clearing of the milk occurs. In few cases acid is produced by the fermentation of lactose and glucose. None of the pathovars coagulate casein. They do not hydrolyze fats nor act upon pectates and do not produce nitrite from nitrate. They are indole negative. Many carbohydrates are used in the study of bacteria. Fermentation and acid production can be determined by the behaviour of the organism in the carbohydrate, alcohol or glucoside media. Most of the *X.c.* pathovars are known to produce acid from carbohydrates and sugar derivatives except salicin and dulcitol and rarely from sorbitol, rham-

TABLE 1.—*X. campestris* pathovars recorded (1915-1989) in Puerto Rico.

Pathovar		Host	Disease	Symptoms	Recorded by/year
<i>aracearum</i>	Tanier	<i>Xanthosoma sagittifolium</i> (L.) Schott	Leaf spot or bacterial spot	Water-soaked lesions surrounded by chlorotic halo. Coalescing lesions toward the central vein	Cortés-Monllor, 1986 (9)
<i>begonia</i>	Begonia	<i>Begonia decandra</i> Pavón	Bacterial leaf spot	Water-soaked spots, marginal necrosis, wilt and death.	Cortés-Monllor, 1971 <sup>1</sup>
<i>cajani</i>	Pigeon pea	<i>Cajanus cajan</i> (L.) Millsp. cv. Kaki	Leaf spot	Small leaf spots, necrotic center and chlorotic halo	Cortés-Monllor and Ruiz, 1986 (11)
<i>campestris</i>	Cabbage	<i>Brassica oleracea</i> L. cv. Market price	Black rot	Marginal necrosis, black veins and exudate	Thomas, 1917 (24)
	Tanier	<i>X. sagittifolium</i> L. Schott	Marginal necrosis or bacterial necrotic margin	Yellow rim on edge of leaf, turning necrotic, dry, spreading toward the central vein.	Cortés-Monllor, 1988 <sup>1</sup>
	Turnip	<i>Brassica rapa</i> L.	Black rot	Stunting and blackening of leaf veins, dry rot of fleshy root	Stevenson, 1917 (22)
<i>dieffenbachiae</i>	Anthurium	<i>Anthurium andreanum</i> L. cv. Nitta	Bacterial wilt (Bacterial blight)	Yellow-brown spots, latter necrotic with chlorotic halo, defoliation, stem necrosis, exudate, wilt and death	Sánchez, 1986 (21)
	Dieffenbachia	<i>D. pictae</i> Schott	Leaf spot	Small chlorotic spots on leaves, changing to brownish. Wilt and death of infected leaves may occur	Stevenson, 1939 (22)
<i>glycines</i> ( <i>X. phaseoli</i> var. <i>sojensis</i> )	Soybean	<i>Glycine max</i> (L.) Merr. cvs. Hutton, Hardee, Kanrich, Pelican, Pance, PI 196-193, 972 A-45, and 317-334B	Bacterial pustules	Small leaf spots, narrow chlorotic halo, necrotic center usually cracks	Leypon, 1972 (16)

<i>malvacearum</i>	Cotton	<i>Gossypium hirsutum</i> L.	Bacterial blight	Angular leaf spots, water-soaked, brownish, gummosis of bract and bolls	Cook, 1934 (2)
<i>phaseoli</i> <i>X.c. vignicola</i>	Bean	<i>Phaseolus vulgaris</i> L. cv. Black Valentine cv. Santa Ana	Common blight	Leaf spots with bright yellow margin, necrotic center, stem necrosis and defoliation	Stevenson, 1915 (22) Vakili, 1975 (25) Cortés-Monllor, 1972 <sup>1</sup>
<i>rubrilineans</i>	Sugarcane	<i>Saccharum officinarum</i> L.	Red stripe	Long, narrow red stripes on leaves, some coalescing forming broad bands. Top rot may occur	Cook, 1929 (2)
<i>rubrisubalbicans</i>			Mottle stripe	Pale red stripes on leaves, some coalescing to form reddish and white bands	Cook, 1932 (2)
<i>stewartii</i>	Maize	<i>Zea mays</i> L.	Bacterial wilt	Pale yellow stripes on leaves, dwarfing, wilt and death	Cook, 1925 (2)
<i>syngonii</i>	Nephthytis	<i>Syngonium podophyllum</i> Schott	Leaf blight	Dry necrotic lesion, surrounded by chlorotic halo	Cortés-Monllor, 1989 (10)
<i>vascolorum</i>	Sugarcane	<i>Saccharum officinarum</i> L. cvs. H-109, Rayada, Cristalina and B-4362	Gummosis (Gummy disease)	Vertical yellowish leaf stripe scattered brownish spots, exudate, dwarfing and decay	Matz, 1919 (2)
<i>vesicatoria</i>	Pepper	<i>Capsicum annum</i> L. cv. Cubanelle, Blanco del País	Bacterial scab (Bacterial pustule)	Small necrotic leaf spots, chlorotic halo. Fruit lesions are concave with central pustule	Cook, 1931 (2)
	Tomato	<i>Lycopersicon esculentum</i> Mill. cv. Duke, Manalucie			Perez and Cortés-Monllor 1960 (18)

<sup>1</sup>Reported herein for the first time.



<u>Localities</u>	<u>Cultivar</u>	<u>Pathovars</u>
1- Río Piedras	Bean Cabbage Syngonium	<u>phaseoli</u> <u>campestris</u> <u>syngonii</u>
2- Isabela	Tanier  Soybean Pepper Tomato Beans	<u>aracearum</u> <u>campestris</u> <u>glycines</u> <u>vesicatoria</u> <u>vesicatoria</u> <u>phaseoli</u>
3- Añasco	Tanier	<u>aracearum</u>
4- Dorado	Pepper Soybean	<u>vesicatoria</u> <u>glycines</u>
5- Naranjito	Anthurium Dieffenbachiae spp Begonia	<u>dieffenbachiae</u> <u>dieffenbachiae</u> <u>begonia</u>
6- Adjuntas	Anthurium Soybean	<u>dieffenbachiae</u> <u>glycines</u>
7- Barranquitas	Cabbage	<u>campestris</u>
8- Corozel	Soybean	<u>glycines</u>
9- Jayuya	Tomato	<u>vesicatoria</u>
10- Mayagüez	Anthurium Bean	<u>dieffenbachiae</u> <u>phaseoli</u>
11- Santa Isabel	Cabbage Pigeon pea Pepper Tomato	<u>campestris</u> <u>cajani</u> <u>vesicatoria</u> <u>vesicatoria</u>

PLATE 4.—Map of Puerto Rico indicating localities of cultivars affected by *X. campestris* pathovars.

nose, and inositol. Tables 2, 3, and 4 present the main biochemical reactions of some *X.c.* pathovars isolated in Puerto Rico from various sources.



TABLE 2.—Biological reactions of *X. campestris* pathovars isolated from various sources in Puerto Rico

Test	Pathovar/Source											
	<i>aracearum</i> Tanier	<i>begonia</i> Begonia	<i>cujani</i> Pigeon pea	<i>campestris</i> Cabbage	<i>tanier</i> Tanier	<i>dieffenbachia</i> Anthurium	<i>glycines</i> Soybean	<i>phaseoli</i> Bean	<i>vascolorum</i> Sugarcane	<i>vesicatoria</i> Pepper	<i>tomato</i> Tomato	<i>syngonii</i> Nephthytis
Indole production	-	-	-	-	-	-	-	-	-	-	-	-
Methyl-Red	-	-	-	-	-	-	-	-	-	-	-	-
Voges-Proskauer	-	-	-	-	-	-	-	-	-	-	-	-
Nitrite from nitrate	-	-	-	-	+	-	v	-	-	-	v	-
Ammonia production	+	-	+	+	+	-	+	v	-	-	+	+
Urea production	-	-	-	-	-	-	v	v	-	-	-	-
Citrate utilization	-	+	+	+	+	+	+	+	+	+	+	+
Malonate utilization	-	-	+	+	+	+	+	+	v	+	+	-
H <sub>2</sub> S from cysteine and peptone	+	-	+	+	-	+	+	+	+	+	+	+
Liquefaction of yeast pectate	-	-	-	+	-	-	v	-	-	-	-	-
Liquefaction of gelatin	v	+	+	v	+	+	+	v	v	+	+	+
Lypolysis	-	-	+	v	-	-	v	-	-	-	v	-
Esculin hydrolysis	+	.	+	+	+	+	+	+	+	+	+	+
Salt tolerance 2-10%	2%	.	3%	2%	2%	2%	3%	3%	3%	3%	3%	2%
Hydrolysis of starch	+	+	+	+	+	+	+	+	v	v	v	.
Catalase	+	.	+	+	+	+	+	+	.	+	+	+
Oxidase	-	.	-	-	-	-	-	-	.	-	-	-

Data: + = positive      v = variable  
 - = negative      . = no record

TABLE 3. Action on litmus milk by *X. campestris* pathovars<sup>1</sup>

Pathovar/Source	Litmus Milk
<i>aracearum</i> Tanier	Alkaline, slow peptonization
<i>begonia</i> Begonia	Yellow pellicle and ring at top, slow clearing, yellowish liquid
<i>cajani</i> Pigeon pea	Acid top, casein digested
<i>campestris</i> Tanier & Cabbage	Cabbage: Slightly alkaline at top, yellowish ring, slow peptonization, clearing Tanier: acid, white and clearing
<i>dieffenbachia</i> Anthurium	Slightly acid top, clearing, yellow sediment
<i>glycines</i> Soybean	Alkaline, slow peptonization
<i>phaseoli</i> Bean	Alkaline, slow peptonization
<i>vasculorum</i> Sugarcane	Alkaline, yellow ring, peptonization
<i>vesicatoria</i> Pepper & tomato	Pepper: Alkaline, yellow ring, peptonization, clearing, and yellow sediment Tomato: Alkaline, peptonization, clearing
<i>syngonii</i> Nephthytis	Clearing on top, yellowish ring, peptonization

<sup>1</sup>Reading recorded 1 month after inoculation.

#### GEOGRAPHICAL DISTRIBUTION

Puerto Rico is located in the tropical zone. Mean temperature is 76.5°F; mean annual rainfall, between 102 cm in the South Coast and 382 cm in the Eastern Interior (20). In the tropics, moisture is more critical for agricultural purposes because temperature changes throughout the year are insignificant (7). Tropical climate is one of the most compatible factors for bacterial diseases. Favored by this factor, *X.c.* pathogens are widespread in Puerto Rico.

Sugarcane has been an important crop in Puerto Rico since 1548. When gummosis was first reported in 1919 (2), the affected plantations were in Caguas (East-central region) and Canóvanas (Northern slope), Cayey (Eastern interior) and Morovis (Western interior). Varieties planted (Otaheite, Rayada, P.R. 491 and Cristalina) were all susceptible. Attempts to find resistant varieties were not successful. The disease spread to Toa Baja (north coast), Trujillo Alto, Isabela, Añasco,

TABLE 4.—Acid production\* from carbohydrates by pathovars of *X. campestris* isolated from different plant species in Puerto Rico

Carbohydrate	Pathovar/Source											
	<i>aracearum</i> Tanier	<i>begonia</i> Begonia	<i>cajani</i> Pigeon pea	<i>campestris</i> Cabbage	<i>tanier</i> Tanier	<i>dieffenbachia</i> Anthurium	<i>glycines</i> Soybean	<i>phaseoli</i> Bean	<i>vasculorum</i> Sugarcane	<i>vesicatoria</i> Pepper	<i>tomato</i> Tomato	<i>syngonii</i> Nephthytis
Arabinose	+	-	.	+	+	+	+	+	+	+	+	+
Cellulose	+	.	.	v	+	+	.	-	.	+	.	+
Dextrine	.	-	.	+	.	.	+	+	+	+	-	.
Dulcitol	.	-	.	-	.	.	-	-	-	-	-	-
Galactose	.	-	.	+	.	.	+	+	+	+	+	+
Glucose	+	.	+	+	+	+	+	+	+	+	+	+
Glycerol	+	+	.	-	.	-	-	-	-	-	-	.
Inositol	+	.	.	-	-	v	.	-	.	-	-	v
Lactose	-	-	.	v	-	+	+	+	+	+	-	-
Levulose	+	+	.	+	+	+	.	+	+	+	+	.
Maltose	+	-	+	+	+	+	-	v	+	v	v	v
Mannite	-	-	.	v	-	-	-	-	-	v	v	-
Mannose	+	-	.	+	+	+	+	v	+	v	.	+
Raffinose	.	-	.	+	.	.	+	+	-	+	v	.
Rhamnose	-	+	.	-	-	v	-	+	-	+	-	-
Salicine	-	-	-	-	-	v	-	-	-	-	-	-
Sorbitol	v	-	.	-	-	v	-	-	-	-	v	-
Sucrose	+	-	+	+	+	+	+	+	+	+	v	+
Trehalose	+	.	.	+	+	+	+	+	.	+	.	+
Xylose	+	+	.	+	+	+	+	+	+	+	+	-
		slow			slow			slow				

Data: \* 30 days observation  
 + = positive  
 - = negative  
 V = variable  
 . = no record

Bayamón, and Fajardo (northern slope), Río Grande (eastern interior), Adjuntas, Guayanilla and Mayagüez (western interior), Humacao and Naguabo (southern slope), Vieques (off-shore island) and to Coamo, Salinas and Yauco (south coast). Other varieties planted (B-376, B-356, Calancana, Cavengerie, D-109, and Yellow Caledonia) were also susceptible. Plate 3 shows the distribution of *X.c. vasculorum* in sugarcane growing areas. Gummosis was a serious disease for many years but lately it has not been detected probably because of the elimination of many sugarcane plantations. Many other sugarcane-growing countries have not reported the disease; in some it is presumed to have been controlled with the use of resistant varieties.

Minor crops and vegetables, i.e., beans, soybeans, cabbage, turnip, pigeon peas, taniers, tomatoes, and peppers, have been affected by *X. campestris* pathovars in most of the areas planted (table 5 and plate 4).

*X.c. vesicatoria*, which causes bacterial spot or scab in tomatoes and peppers (plate 4), leads to great losses of fruit (8). The organism has been isolated from affected plantations on the northern coast

TABLE 5.—Diseased plant material collected in different locations and annual rainfall distribution in Puerto Rico.<sup>1</sup>

Location	Average Annual rainfall	Samples collected (after 1960)
North Coast Dorado	less than 165 cm	Soybeans, pepper
Northern Slope Río Piedras Isabela  Añasco	152 to 178 cm	Cabbage, beans, syngonium Beans, soybeans, pepper, tomato and tanier Tanier
Eastern Interior Naranjito	178 to 382 cm	Anthurium, begonia, <i>D. pictae</i>
Western Interior Adjuntas Barranquitas Corozal Jayuya	178 to 254 cm	Soybeans, anthurium Cabbage Soybeans Tomatoes
West Coast Mayaguez	102 to 178 cm	Anthurium, beans
South Coast Santa Isabel	less than 102 cm	Pigeon pea, cabbage, pepper, and tomatoes

<sup>1</sup>See Reference 20.



(Dorado), northern slope (Isabela), western interior (Jayuya) and the southern coast (Santa Isabel). Pepper varieties Cubanelle and Blanco del País and tomato varieties Manalucie and Duke are among the cultivars most affected.

Pathogens *X.c. dieffenbachiae* and *X.c. begoniae* have been isolated from Anthurium and Begonia plants, respectively. These ornamentals are mostly grown in the eastern and western interiors, where elevation ranges from 120 to 450 m and annual rainfall from 178 to 382 cm, adequate conditions for ornamental horticulture. The disease is damaging in nurseries where plants are grown under high humidity. Bacterial isolates were identified from nurseries in Adjuntas (western interior) and Naranjito (eastern interior) and in Mayagüez (western region).

Table 5 presents data of samples collected in dispersed locations (plate 4) where rainfall patterns are very variable (2).

#### ANTIBIOTIC SENSITIVITY

Laboratory test used to determine resistance or susceptibility of the organisms to different antibiotics was performed as stated in Difco (12). Close similarity in the reactions of the *X.c.* pathovars to the various antibiotics tested was observed. *X.c. vesicatoria* exhibits slight differences in its reaction susceptibility in tomato and pepper strains. Tomato strains show a larger zone of growth inhibition around the antibiotics tested than the pepper strains at the same concentrations. Bradbury (5) states that *X.c. vasculorum* has been separated into various types by its sensitivity to some antibiotics as this species is very heterogeneous. All local isolates tested were susceptible to aureomycin, chloramphenicol, erythromycin (except *X.c. aracearum* and *phaseoli*, which were moderately susceptible), novobiocin and tetracycline. They were moderately susceptible to polymyxin B and resistant to neomycin and penicillin G. Various degrees of sensitivity to streptomycin and bacitracin were noted among the pathovars.

Table 6 presents results obtained in the *in vitro* susceptibility test of the *X.c.* pathovars to different antimicrobial agents tested.

#### DISEASE CONTROL

Bacterial diseases of tropical plants and perennial crops present year-round problems because of the prolonged association of the organisms with the host. A few bacterial diseases can be controlled under field conditions; however, infected plants are not cured.

Little work has been done on the control of some bacterial diseases. Disease intensity is variable within the area of occurrence. Areas with climatic conditions unfavorable to the disease can be selected for farming, although more research is needed on how environmental conditions can

TABLE 6.—Susceptibility of *X.c.* pathovars to different antibiotics<sup>1</sup>

Antibiotic		Concentration per disk	<i>X.c. pathovars</i>							
			<i>aracearum</i>	<i>cajani</i>	<i>campestris</i>	<i>dieffenbachiae</i>	<i>phaseoli</i>	<i>syngonium</i>	<i>vesicatoria</i>	
			Tanier	pigeon pea	Tanier	Anthurium	Bean	Nephthytis	Pepper	Tomato
Tetracycline	(TE <sub>30</sub> )	30 ug	S	S	S	S	S	S	S	S
Aureomycin	(A <sub>30</sub> )	30 ug	S	S	S	S	S	S	S	S
Chloramphenicol	(C <sub>30</sub> )	30 ug	S	S	S	S	S	S	S	S
Streptomycin	(S <sub>10</sub> )	10 ug	R	MS	R	R	S	S	S	MS
Polymyxin B	(PB <sub>300</sub> )	300 units	MS	MS	MS	MS	MS	MS	MS	MS
Neomycin	(N <sub>30</sub> )	30 ug	R	R	R	R	R	R	R	R
Erythromycin	(E <sub>15</sub> )	15 ug	MS	S	S	S	MS	S	S	R
Bacitracin	(B <sub>10</sub> )	10 units	R	S	R	R	MS	S	R	MS
Novobiocin	(NB <sub>30</sub> )	30 ug	S	S	S	S	S	S	S	S
Penicillin	(P <sub>10</sub> )	10 units	R	R	R	R	R	R	R	R

<sup>1</sup>Results reported according to the interpretation of the zone diameters stated in table I in Difco Manual (12).

S = Susceptible

MS = Moderate susceptible

R = Resistant

modify or regulate the host-pathogen interaction. Environmental factors (temperature and moisture) can be controlled in nurseries and greenhouses to produce pathogen-free seeds and planting material. Incidence of bacterial blight of beans (*X.c. phaseoli*), black rot of cabbage (*X.c. campestris*) and bacterial scab of tomato (*X.c. vesicatoria*) is greatly reduced by using disease free seeds. These are seed-borne pathogens. Farmers unaware of this fact contribute to the spread of these diseases. Once in the field, the organism spreads with the help of wind and rain. Cultural practices can diminish severity and incidence of most ornamental foliage diseases (leaf blight of begonia, anthurium, dieffenbachia, etc.) caused by *X.c.* pathovars. Control measures recommended include maintenance of field sanitation and destruction of infected material. Use of contaminated tools and of overhead sprinkling should be avoided. Chemical treatment has been recommended for control of leaf spots in beans, peppers and tomatoes (8, 16, 26) affected with *X.c. phaseoli* and *X.c. vesicatoria*, respectively, and for anthurium and dieffenbachia species affected with *X.c. dieffenbachiae* (21). Spraying with copper compounds and antibiotics can minimize the disease, although most of the bactericides are not effective in controlling systemic pathogens. Research is needed in this area in order to develop new bactericides equally safe for consumers, plants and farmers. Recently, specific nutrients have been employed successfully for controlling some *Xanthomonas* (10).

Disease control of many *X.c.* pathovars through the use of resistant varieties is not yet available. As stated before, gummosis has almost disappeared from sugarcane plantings through the use of varieties resis-

TABLE 7.—*Bacterial diseases caused by X.c. pathovars that had not been recorded in Puerto Rico*

<i>X.c.</i> pathovar	Disease	Cultivar
<i>alfalfae</i>	Leaf spot	Alfalfa (Lucerne)
<i>cassava</i> (27)	Bacterial blight	Cassava
<i>celebensis</i>	Blood disease	Plantains & bananas
<i>citri</i> (27)	Citrus canker	Citrus
<i>cucurbitae</i>	Bacterial spot	Cucurbits
<i>holcicola</i>	Bacterial streak	Maize & sorghum
<i>mangiferae-indicae</i> (27)	Black spot	Mango
<i>manihotis</i>	Bacterial blight	Cassava
<i>oryzicola</i>	Bacterial blight	Rice
<i>pelargonii</i>	Bacterial blight (Bacterial wilt)	Geranium
<i>poinsettiaeicola</i>	Leaf spot	Poinsettia
<i>vitians</i>	Dry leaf spot Head rot	Lettuce
<i>zinniae</i>	Bacterial leaf spot	Zinnia

tant to *X.c. vasculorum*. In Puerto Rico, resistance to bacterial scab of tomato (*X.c. vesicatoria*) and to bacterial bean blight (*X.c. phaseoli*) has been studied (26, 27). Recently high levels of resistance to *X.c. phaseoli* (bacterial bean blight) have been reported (29).

Quarantine precautions are very important in avoiding propagation of pathogens introduced through seeds or vegetative material. Imported plant material should be submitted to inspection, hot water treatment (where appropriate) and observation. Some bacterial diseases that have not been detected in Puerto Rico occur in areas that are dangerously close to the island. Among these are citrus canker and bacterial blight of cassava affected by *X.c. citri* and *X.c. manihotis*, respectively (14). During the years that rice was grown on the eastern and western coasts of Puerto Rico, bacterial blight (*X.c. oryzae*) was not detected. Table 7 lists some of the *X.c.* pathovars that could be introduced and would infect many of the crops grown in Puerto Rico (8).

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