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WHY AND HOW TO DISTINGUISH THE *PSEUDOMONAS SOLACEARUM* STRAINS, CAUSAL AGENT OF THE BACTERIAL WILT OF SOLANACEOUS AND MUSACEOUS CROPS IN THE CARIBBEAN ZONE

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Bacterial wilt affects solanaceous and musaceous crops in all the countries of the Caribbean region. It is however difficult to estimate exactly the extent of losses caused by this disease. In some territories, the pathogen is so virulent throughout the year that susceptible crops are difficult to grow or have entirely disappeared. In other countries the bacterium is virulent only during certain months of the year (wet season) and susceptible crops are grown during the period unfavourable to the expression of bacterial virulence (dry season).

The extent of losses caused by bacterial wilt is always a direct function of bacterial virulence. If the virulence of the bacterium is subjected to the influence of the ecological factors (climate, soil, host-plant) it is at first conditioned by the nature of the bacterial population, i.e. by the nature of strains in the soil. Every strain has a definite virulence power. It is, therefore, useful to distinguish between one strain and another.

The purpose of this work is to describe certain specific characters which allow you to recognize the *Pseudomonas solanacearum* strains in the Caribbean zone. Of the specific characters only pathotype, biotype and serotype will be studied.

PATHOTYPE

The Pathotype of a bacterial strain is the pathogenicity possessed by that strain towards one or several host plants.

The pathotype gives only the relative value of the pathogenic potential of the strain.

When the pathotype of one isolate is studied, several difficulties arise since the quantity and the quality of the inoculum is dependent on the inoculation methods used for the host-plants.

1. INOCULUM

The isolate must possess its highest virulence potential

To obtain the highest level of virulence of an isolate it is necessary to make the isolation on a selective medium (as KELMAN'S medium) (11) especially since the ratio of virulent to avirulent cells in inoculum influences the severity of bacterial wilt (2).

The highest level of virulence must be maintained before and during the inoculation of the host plant

The best means of maintaining virulence of the strains is to keep the isolate under sterile distilled water (3, 4, 5).

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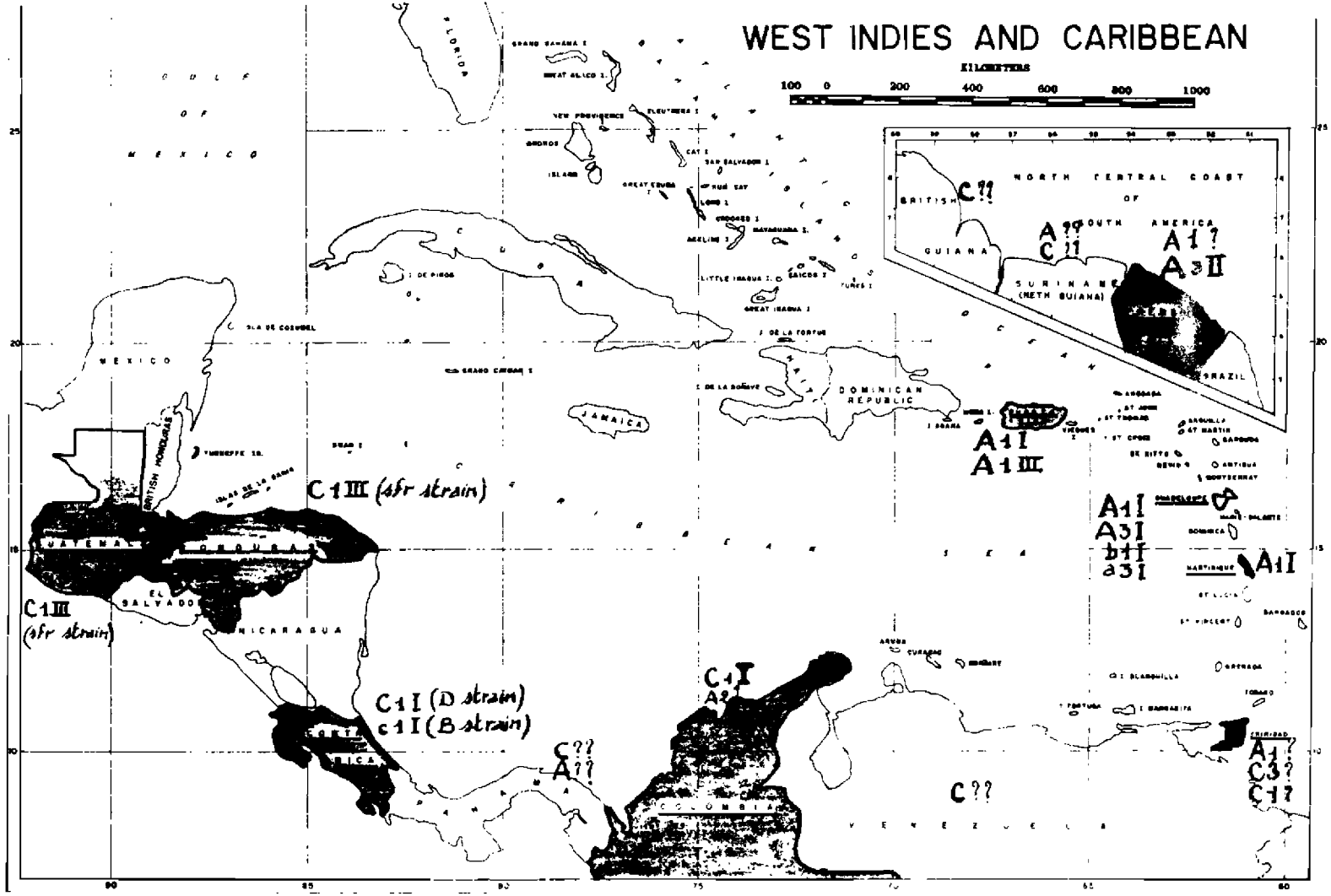
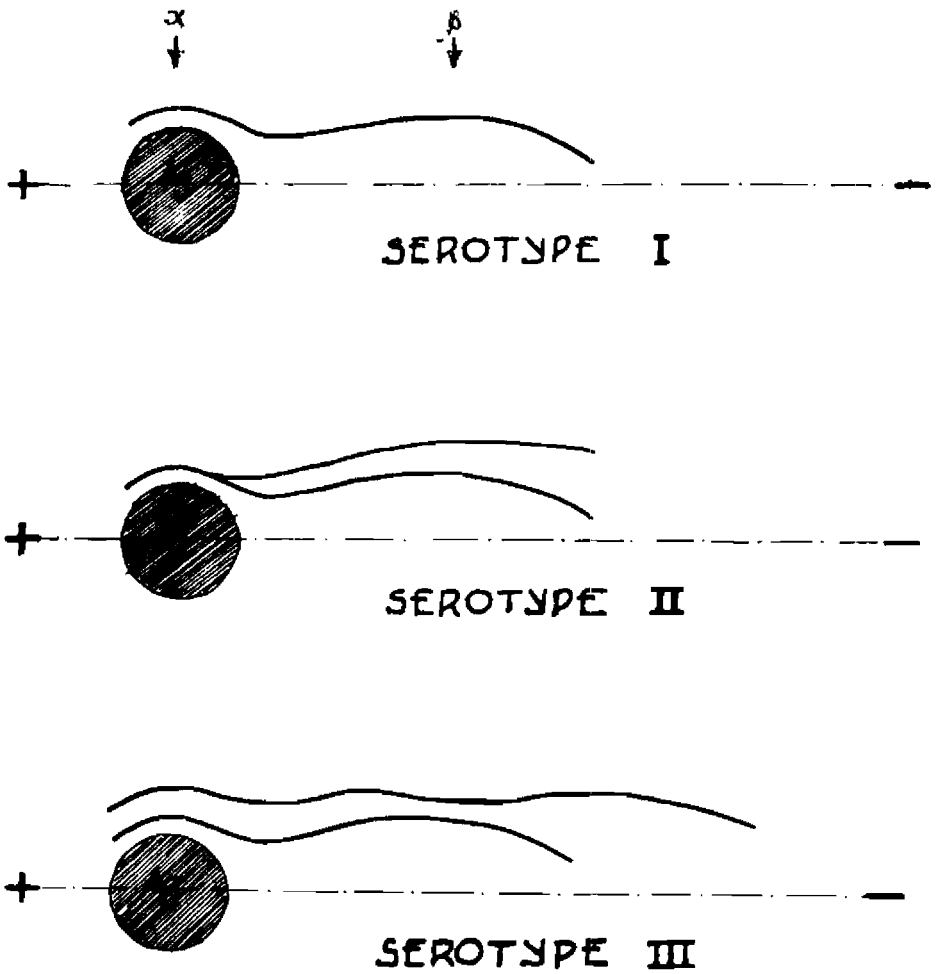
CURRENT STATUS OF THE DISTRIBUTION OF THE PSEUDOMONAS SOLANACEARUM STRAINS.



SCHEMATIC REPRESENTATION OF PRECIPITATION LINES PATTERNS

obtained by immunoeclectrophoresis.



Ag: Hole, cut in the agar, filled with somatic "O" antigen solution extracted from a strain of Pseudomonas solanacearum E.F. Sm.

2. INOCULATION METHODS

The root-inoculation and stem-inoculation methods have been described by WINSTEAD AND KELMAN (6).

3. HOST-PLANT RANGE

Four groups of host-plants A, B, C, D are proposed, as shown in Table 1. According to the observed symptoms in these host plants, the pathotype of the isolate may be restricted to one group or may belong to several groups.

If the symptoms are severe ("fast wilt"), the designated letters for the pathotype are A, B, C, D. But if the symptoms are mild (slow wilt) the designated letters are respectively a, b, c, d.

The isolate is classified as to pathotype only when it can produce in the group, symptoms of wilt. For instance, isolates from Guadeloupe belong to the three pathotypes A, a and b (*see* Table 2).

II. BIOTYPE

According to HAYWARD (7), the biotype or biochemical type is the power of a *Pseudomonas solanacearum* strain to utilize or not:

—the three disaccharides (maltose, lactose, cellobiose).

—the three hexose—alcohols (mannitol, sorbitol, dulcitol).

The method (7) allows us to distinguish four biochemical types according as the Carbohydrate oxidation is positive or negative.

TABLE 1

BIOTYPE / CARBOHYDRATE	1	2	3	4
MALTOSE	−	+	+	−
LACTOSE	−	+	+	−
CELLOBIOSE ...	−	+	+	−
MANNITOL ...	−	−	+	+
SORBITOL	−	−	+	+
DULCITOL	−	−	+	+

The 23 strains of the Caribbean zone belong to *biotype* 1 or *biotype* 3. (*see* Table 3). *There is no direct relationship between the pathotype and the biotype.*

Isolates from PUERTO RICO and MARTINIQUE have the pathotype A but COSTA RICAN and HONDURAN isolates have the pathotype C. However, all these isolates belong to the same biotype 1. The homogeneity inside of some territories is remarkable. In PUERTO RICO, all the isolates from tomato belong to the same biotype 1. In MARTINIQUE, all the isolates from both tomato and eggplant belong to biotype 1. The possibility exists of finding in the same territory two biotypes as in GUADELOUPE where biotype 1 and biotype 3 are present.

III. SEROTYPE

For *Pseudomonas solanacearum*, identities or non-identities among the strains of the bacterium could not be clearly definited (8, 9, 10, 11, 12, 13) and the validity of the notion of serotype or serological type remained uncertain. This was due especially to the lack of sharpness of techniques used in the serological analysis of the *Pseudomonas solanacearum* antigens.

In a previous work (14) it was suggested that the Immunoelectrophoretic analysis of Somatic "O" Antigens could permit the identification of one strain from another.

The 23 Somatic "O" antigens of the strains from the Caribbean zone were extracted and studied.

One type of precipitating antiserum made from the "GUA To 4" strain permits the identification of only 3 different patterns among these 23 antigens.

Serotype I (*see* schematic diagram)

The "GUA To 4" antiserum precipitates its somatic corresponding antigen (extracted from the GUA To 4 isolate) according to two arcs a and B.

This pattern is the most common. Twenty somatic antigens precipitate according to this pattern:

PUERTO RICO	PR 65	MARTINIQUE	MAR To 1
	PR 69		MAR Au 1
	PR 81		MAR Au 2
	PR 211 B		MAR Au 3
	PR 212		
GUADELOUPE	GAU To 1	COSTA RICA	B 139–B
	GUA To 2		B
	GUA To 3		D
	GAU Pt 1		
	GUA Ta 1		
	GUA Ta 2		
	GUA To 4	COLOMBIE	H 249

Serotype II (*see* schematic diagram, Figure 1)

This pattern includes not only the two arcs A and B but a supplementary arc, originating from the A arc with the same shape and length as the B arc.

Only one isolate belongs to this pattern:

FRENCH GUIANA (GUY TO 1)

Serotype III (*see* schematic diagram)

In this pattern, three specific precipiting bands are joined to form a continuous curve, located above the A and B arcs.

Two somatic antigens give this pattern:

HONDURAS (SFR
PUERTO RICO (PR 80.

The specific characters of the *Pseudomonas solanacearum* strains are summarized in Table 3.

The geographical distribution of these strains in the Caribbean zone as seen in the attached map shows that the pathogen is uniformly distributed, but that the pathotype C (virulent strain for the Musaceous crops) is rather continental: GUATEMALA, HONDURAS, COSTA RICA, PANAMA, COLOMBIA, VENEZUELA, BRITISH GUIANA (except for its existence in TRINIDAD which is close to the VENEZUELAN Mainland).

It is very likely that the pathotype A (which is virulent for Tomato, Potato, Eggplant and Pepper) is present in all territories of the Caribbean. Its presence could explain the weak development of the Solanaceous vegetables in this zone.

The lack of biotype 4 and the infrequency of biotype 2 (only present in COLOMBIA) are noticeable.

A more comprehensive study of the serotypes is in progress and will lead to a better understanding of the origin, distribution and properties of some *Pseudomonas solanacearum* strains.

TABLE 1

Host Plants Groups for the determination of Pathotypes of *Pseudomonas Solanacearum* Isolates

A (*a*) (Vegetable Solanaceous)	B (*b*) (Tobacco Group)	C (*c*) (Musaceous)	D (*d*)
TOMATO (*Lycopersicon esculentum Mill*)	TOBACCO (*Nicotiana tabacum L.*)	BANANA (*Musa. spp.*)	PEANUT (*Arachis hypogaea L.*)
POTATO (*Solanum tuberosum L.*)		HELICONIA *caribea*	SESAME (*Sesame indicum L.*)
EGGPLANT ((*Solanum melongena L.*)		HELICONIA *psittacorum*	GINGER (*Zingiber officinale Rosc.*)
PEPPER (*Capsicum frutescens L.*)		HELICONIA *spp.*	
		STRELITZIA *spp.*	

TABLE 2

Pathotypes of the isolates of *Pseudomonas Solanaccarum* E. F. Sm. in Guadeloupe

ISOLATES from / HOST-PLANT GROUP	TOMATO (*fast wilt*)	TOMATO (*slow wilt*)	TOBACCO (*slow wilt*)
A (*a*)	*	†	‡
B (*b*)	§	§	†
C (*c*)	§	§	§
D (*d*)	+	§	*
PATHOTYPE	A	a	b

Degrees of virulence: * strong (fast wilt).
† medium (slow wilt).
‡ weak (no wilt).
§ avirulence.

TABLE 3

Specifical Characters of the Pseudomonas Solanacearum Strains in the Caribbean Zone

TERRITORY OF ORIGINE	AREA OF SAMPLING	HOST-PLANT	NUMBER OF THE ISOLATE	AUTHORS, DATE OF ISOLATION	PATHO-TYPE	BIOTYPE	SERO-TYPE
PUERTO-RICO	?	TOMATO	PR 65	J. E. PEREZ and A. O. MONLLOR 1962 (1958)	A	1	I
	?	TOMATO	PR 69	,, ,,	A	1	I
	?	TOMATO	PR 80	,, ,,	A	1	III
	?	TOMATO	PR 81	,, ,, 1963 (1959)	A	1	I
	?	TOMATO	PR 211B	,, ,, ?	A	1	I
	?	TOMATO	PR 212	,, ,, ?	A	1	I
GUADELOUPE	Duclos	TOMATO	GUA TO 1	B. DIGAT 1965	A	3	I
	Longueteau	TOMATO	GUA TO 2	A. ESCUDIE 1964	A	1	I
	Vx Habitants	TOMATO	GUA TO 3	B. DIGAT 1966	A	1	I
	Pte Noire	POTATO	GUA PT 1	,, ,, 1965	A	1	I
	Capesterre	TOBACCO	GUA TA 1	,, ,, 1967	b	1	I
	Roujol	TOBACCO	GUA TA 2	,, ,, 1967	b	1	I
	Beauport	TOMATO	GUA TO 4	,, ,, 1965	a	3	I
MARTINIQUE	Basse-Pointe	TOMATO	MAR TO 1	B. DIGAT 1965	A	1	I
	Basse-Pointe	EGGPLANT	MAR AU 1	,, ,, 1967	A	1	I
	Saint-Pierre	EGGPLANT	MAR AU 2	,, ,, 1967	A	1	I
	Morne-Rouge	EGGPLANT	MAR AU 3	,, ,, 1967	A	1	I
TRINIDAD	?	MUSA sp.	a 446 (TRI BA 1)	A. C. HAYWARD 1957	C	1	?
	La REUNION Trinidad Government Cocoa propagation unit	MUSA "Mysore"	TRI BA 2	B. DIGAT 1968	C	3	?
	Las Lomas	MUSA "Moko Fig"	TRI BA 3	,, ,,	C	3	?
	Matura estate	MUSA "Giant Cavendish"	TRI BA 4	,, ,,	C	3	?
	Matura estate	MUSA "Lacatan"	,, ,,		C	3	?
	Sangre Grande "El Reposo" Station	MUSA "Horse plantain"	TRI BA 5	,, ,,	C	1	?
	Sangre Grande Sainte-Marie	MUSA "Mysore"	TRI BA 6	,, ,,	C	1	?
	?	TOMATO	a 616 (TRI TO 1)	W. J. DOWSON 1959 J. A. SPENCE 1957	A	1	?
	Aranguez Market Garden Estate	TOMATO	TRI TO 2	B. DIGAT 1968	A	1	?
	Caura Valley	TOMATO	TRI TO 3	,, ,, 1968	A	1	?
	St. Helena Plarco EGG-PLANT	TOMATO	TRI TO 4	,, ,, 1968	A	1	?
	Sangre Grande "El Reposo" Station	TOMATO	TRI TO 5	,, ,, 1968	A	1	?
FRENCH GUIANA	Matoury (Sicama)	TOMATO	GUY TO 1	H. FLOCH and B. DIGAT 1965 1968	A	3	II
	Cayenne Suzini	TOMATO	GUY TO 2	B. DIGAT 1968	A	1	?
	Kourou (Seac)	TOMATO	GUY TO 3	B. DIGAT 1968	A	?	?
COSTA RICA	Coto Valley	BANANA	B. 139—B	I. W. BUDDENHAGEN 1959	C	1	I
		BANANA	B	,, ,, ,, ,,	C	1	I
		HELICONIA ssp	D	,, ,, ,, ,,	C	1	I
HONDURAS	Ulua Valley	BANANA	SFR	I. W. BUDDENHAGEN 1961	C	I	III
COLOMBIA	?	HELICONIA CARIBEA	H 249	I. W. BUDDENHAGEN 1960	C	1	I

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