



HAL
open science

Variations among strains of *Xanthomonas Campestris* isolated from Citrus distinguished with their sensitivity to antibiotics

Christian Vernière, Olivier Pruvost, C. Dubois, X. Perrier, A. Couteau, J. Luisetti

► To cite this version:

Christian Vernière, Olivier Pruvost, C. Dubois, X. Perrier, A. Couteau, et al.. Variations among strains of *Xanthomonas Campestris* isolated from Citrus distinguished with their sensitivity to antibiotics. 8. International Conference on Plant Pathogenic Bacteria, Jun 1994, Versailles, France. 1027 p. hal-02778944

HAL Id: hal-02778944

<https://hal.inrae.fr/hal-02778944v1>

Submitted on 4 Jun 2020

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Variations among the strains of *Xanthomonas* isolated from citrus in the sensitivity to antibiotics

C. VERNIERE, O. PRUVOST, C. DUBOIS*, A. COUTEAU AND J. LUISETTI**

CIRAD/IRFA, Laboratoire de Phytopathologie,
BP 180, 97455 Saint Pierre Cedex, Réunion, France

* CIRAD/IRFA, Service de Biométrie,
BP 5035, 34032 Montpellier Cedex, France

** INRA, Station de Pathologie végétale,
BP 57, 49071 Beaucauzé Cedex, France

ABSTRACT

65 strains representative of the different pathotypes of *X. campestris* pv. *citri*, the agent of Citrus Bacterial Canker Disease (CBCD), and six strains of *Xanthomonas* responsible for Citrus Bacterial Spot Disease (CBSD) were studied for their *in vitro* sensitivity to 63 antibiotics using a disc test.

All the strains were similarly resistant to four antibiotics, and a variability in the response was observed with the others. Hierarchical cluster analysis of the diameters of zone on inhibition revealed differences between strains of Mascareignes was distinguished. These strains shared a higher resistance to penicillins and cephalosporins. Strains of pathotypes B and D were clearly related, except one strain of the B type. Strains from CBS and strains of the pathotype C exhibited some variability.

These data allowed the selection of an appropriate pool of antibiotics to compose a semi-selective medium for the detection of *Xanthomonas* strains of CBCD and CBSD.

KEYWORDS

Xanthomonas campestris, Citrus bacterial canker disease, Citrus bacterial spot disease, pathotypes, epidemiological marker, identification.

INTRODUCTION

Strains of *Xanthomonas campestris* are responsible of two bacterial foliar diseases on citrus. *X.c.* pv. *citri* is the causal agent of Citrus bacterial canker disease (CBCD) which is widespread in the tropical area. Four variants of this pathogen (called pathotypes A, B, C and D) were initially described according to their host range and their geographical origin. Recently, in 1984, a new foliar disease, named Citrus bacterial spot disease, was found in Florida.

The causal agent is known as *X.c. pv. citri* pathotype E or *X.c. pv. citrumelo* (GRAHAM & GOTTWALD, 1991 ; STALL & CIVEROLO, 1991).

Different laboratory techniques permitted the characterization of each type and confirmed the differences between these pathotypes (Biochemical tests, serological tests using polyclonal and monoclonal antibodies, RFLP analyses...). However no difference related to pathogenic specification or geographical origin was detected among strains belonging to a same type (ALVAREZ *et al.*, 1991 ; HARTUNG & CIVEROLO, 1989 ; VERNIERE *et al.*, 1991).

We examined the variations in the sensitivity to antibiotics of representative strains of *X. campestris* isolated from citrus trying to improve the similarities or differences between the pathotypes or strains belonging to a same type.

MATERIALS AND METHODS

Sixty five strains of CBCD (53 strains of pathotype A originating from 23 countries, 8 strains of pathotype B, 3 strains of pathotype C and the single D type strain) and six strains of CBSD obtained from freeze-dried cultures were tested.

The sensitivity to 63 antibiotics was carried out using an *in vitro* antimicrobial diffusion test. Six discs (Diagnostics Pasteur - 92430 Marnes La Coquette - France) were applied on plates containing recently poured YPDA medium (yeast extract 7 g, peptone 7 g, dextrose 7 g, agar 15 g) using a distributor. After two days incubation at 28°C, the diameter of inhibition was measured. Each combination was repeated twice.

Numerical analysis of phenotypic features : the characters states were quantitative and analysis was performed using the average diameter as the clustering criterion. Hierarchical cluster analysis was carried out using the minimization of intraclass variance.

RESULTS

All the strains of CBCD and the six strains of CBSD were resistant to Trimethoprim, Clindamycin, Lincomycin and Metronidazole.

Four strains (CFBP2901 / B type, CFBP2910 / E type, CFBP2906 / C type and CFBP2857 / A type) were strongly differentiated from the other pathotypes (Figure 1). CFBP2901 was more sensitive, and a larger diameter using Penicillins and Cephalosporins was observed than those obtained with the other B types (Table 1).

Mascareignes (JJ9-3 from Mauritius and JJ10-3 from Rodriguez) showed a profile similar to that of the other type A strains.

Table 1 : variations of *X. campestris* isolated from citrus in the sensitivity to penicillins and cephalosporins.

Pathotypes		type A (3)	type A Masca (4)	type A Mauritius (JJ9-3)	types B + D(5)	type B (CFBP 2901)	type C (x 3)	type E (CBS) (x 6)
	number of strains tested	(x 37)	(x 15)	(x 8)	(x 8)	(x 8)	(x 3)	(x 6)
PENICILLINS								
ampicillin	a(1)	9,1	6	6	27,6	51	6	16,5
	v(2)	35,8	0		6,1		0	58
amoxycillin	a	9,1	6	6	42,3	55	8,5	17,5
	v	50,5	0		6,7		18,8	115
carbenicillin	a	31,8	6,7	36	41,5	55	12,5	40,2
	v	62,9	2,35		3,8		127	12,2
mezlocillin	a	28,2	15,3	36,5	29,9	40	20,3	30,5
	v	28,3	26,6		7,0		7,6	11,1
ticarcillin	a	30,1	6,06	6	43,9	59	12,5	40,4
	v	54,4	0,06		6,0		108	18,0
CEPHALOSPORINS								
cephalexin / 1	a	6	6	6	6	24	8,0	6,6
	v	0	0		0		12,0	2,6
cefazolin / 1	a	6,07	6,03	6	6	34	7,6	7,6
	v	0,09	0,02		0		8,3	7,0
cefamandole/2	a	19,3	7,4	25	20,5	41	13,0	23,6
	v	30,5	5,5		12,4		57,3	18,8
cefuroxim / 2	a	11,4	6,9	13	19,1	44	12,8	19,0
	v	35,2	3,6		9,03		80,6	66,7
cefotiam / 3	a	8,1	6,1	6	21,4	45	11,2	17
	v	27,2	0,07		6,4		80,1	79,9
cefsulodin	a	29,2	6,9	34,5	35,9	51	17,7	36,2
	v	41,4	9,5		5,7		88,6	13,4

(1) average (mm) of the diameters of inhibition - the diameter of the disc is 6 mm.

(2) variance of the diameters of inhibition.

(3) pathotype A strains, strains from Mascareignes not included.

(4) strains from Mascareignes, strain JJ9-3 not included.

(5) pathotypes B and D, strain CFBP2901 not included.

Hierarchical cluster analysis revealed a highly separated group (index of relative distance ID = 0,818) which included fourteen isolates (on sixteen) from the Mascareignes archipelago (Réunion, Mauritius and Rodriguez islands) and one strain from Oman (JF90-5) (Figure 1). These strains were distinguished from the other A type strains according to the higher resistance exhibited with Carbenicillin, Ticarcillin, Cefamandole and Cefsulodin (Table 1).

