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BACTERIAL BLIGHT (*X. campestris* pv. *dieffenbachiae*) AND
BACTERIAL LEAF SPOT (*Pseudomonas* sp.) OF ANTHURIUM
IN THE FRENCH WEST INDIES

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ABSTRACT

Symptoms of decline appeared on hybrid Anthurium in Guadeloupe and Martinique in 1982. The phytosanitary inquiry realized in 1984 has shown that the decline is parasitic and that two different facies are observed according to the pathogenic bacterium involved. *Xanthomonas campestris* pv. *dieffenbachiae* is the limiting factor of the development of this crop, and *Pseudomonas* sp. is reported as a new pathogen which can cause important losses, particularly during the rainy season. Symptoms observed on naturally infected material, as well as pathogenicity tests, distinguished airborne and soil-borne infections. Host range, biochemical and serological characteristics are reported. Measures of control, which are based on sanitation, are presented.

RESUME

Des symptômes de dépérissement sont apparus sur Anthurium en Guadeloupe et en Martinique en 1982. L'enquête phytosanitaire réalisée en 1984 a montré que ce dépérissement est d'origine parasitaire et peut présenter deux faciès différents selon la bactérie en cause. *Xanthomonas campestris* pv. *dieffenbachiae* est le facteur limitant le développement de la culture; *Pseudomonas* sp., nouvellement décrit, peut causer des dégâts importants, en particulier pendant la saison des pluies. L'observation des symptômes, à la suite d'infections naturelles et d'inoculations, permet de caractériser l'origine, aérienne ou tellurique, de l'inoculum. Le spectre d'hôtes, les caractéristiques biochimiques et sérologiques sont décrits. Les moyens de lutte, qui reposent sur l'application de mesures prophylactiques, sont présentés.

INTRODUCTION

Anthurium andreanum and its hybrid varieties are economically important crops in many tropical countries. The development of anthurium in the French West Indies has great potential for the diversification of agriculture and the improvement of social and economic conditions. In 1982, however, symptoms of decline first appeared. This was true for both *Anthurium* sp. cultivated using traditional methods and hybrid anthuriums which require a high degree of technical know-how. This decline has caused significant losses to commercial anthurium growers and continues to limit the development of this crop in Guadeloupe and Martinique. The results of an investigation into this program indicate that severe damages were caused by both *Xanthomonas campestris* pv. *dieffenbachiae* and *Pseudomonas* sp.

GEOGRAPHICAL DISTRIBUTION

Bacterial blight of the *Anthurium* caused by *Xanthomonas campestris* pv. *dieffenbachiae* has been reported by:

- o Robbs (1960) in Brasil,
- o Hayward (1972), Nishijima and Fujiyama (1985) in Hawaii,
- o Guevara and Debrot (1985) in Venezuela,
- o Knauss (1975) in Florida,
- o Hostachy et al. (1985) in French West Indies.

Decline caused by *Pseudomonas* sp. has only been reported by Hostachy et al. (1985) in Guadeloupe and Martinique.

SYMPTOMS

Bacterial Blight Caused by *Xanthomonas campestris* pv. *dieffenbachiae*

The first sign of disease is the appearance of irregular water-soaked spots of areas on the underside of leaves. Foliar symptoms usually start at--or close to--the leaf margins and occasionally in the center of a leaf (due to a wound). As the disease develops, the water-soaked spots become necrotic and large brown areas surrounded by bright yellow borders appear on both surfaces of the leaves. On the underside and at the edge of the necroses, star shaped, water-soaked spots (1 to 3 mm diameter) are also to be observed. These continue to appear along the major veins and indicate the progression of the bacteria within the vascular system. The disease now enters into its systemic phase.

The multiplication of the bacteria contribute to the yellowing of the lamina of old leaves. On the spathe, brown or black spots, sometimes surrounded by a violet border, can be seen. The leaf petioles or spathe peduncles rot at the point where they meet the stem. A transverse section of the petiole or of the stem reveals a pale yellow bacterial exudate. The final stage of the disease is the death of the plant. The systemic infection can however be produced quite independently of a foliar infection. In this case, bacteria enter the plant via the roots.

Bacterial Leaf Spot Caused by *Pseudomonas* sp.

Early foliar symptoms of bacterial leaf spot of *Anthurium* may be characterized as small, angular, greasy spots on the underside surface near veins and leaf margins, and on spathes. These lesions develop rapidly into resulting large, black necrotic spots which become grey-black on older and distort leaves. Necrotic spots are surrounded by greasy margins and narrow, bright chlorotic halos. High humidity is required to observe bacterial slime oozing from lesion margins. Typical black or brown necrotic spots, surrounded by violet halos, usually appear on spathes. Infections may progress into veins causing a soft rot of infected tissue resulting in the abscission of leaf and spathe peduncle. Symptoms on systemically infected plants display a general yellowing of the entire lamina and typical black, necrotic lesions progressing from the leaf petioles into major veins. In this case petioles may rot at the base or top depending on the rapidity of bacterial colonization. Plants

with systemic or local infections in the stem, petiole, leaves and spathes usually die.

Important points to avoid confusion

<u>Xanthomonas</u>	<u>Pseudomonas</u>
Large yellow border surrounding necrotic areas	Narrow yellow border surrounding necrotic areas
Star-shaped water-soaked spots on the underside of leaves	Star-shaped water-soaked spots absent

INFECTION

Bacterial decline can result from either one or two different modes of infection: air-borne and/or soil-borne.

Air-borne Infection

For both pathogenic bacteria, this type of infection is brought about by water droplets splashing or by contact with contaminated clothing or tools. The bacteria colonize the leaf parenchyme via natural openings such as stomata, hydathodes or wounds. Every diseased leaf is a potential contributor to the spread of the inoculum in the environment (abscission and scattering of bacteria by rain droplets).

Soil-borne infection

Both pathogenic bacteria enter the vascular system via natural openings or wounds. Once there, they multiply rapidly and spread throughout the plant. In the soil, dissemination by water is rare for *Pseudomonas* sp. but very common for *Xanthomonas campestris* pv. *dieffenbachiae*. This is particularly serious in Guadeloupe where production occurs mainly on slopes under high rainfall conditions. Shelter-houses situated downhill of infected areas have therefore an increased chance of becoming contaminated.

ISOLATION

The isolation methods for both bacteria were more or less identical. Tissue was excised from the margins of lesions on infected anthurium petioles, leaves, stems and flowers. Samples were surface-sterilized with alcohol, crushed in sterilized distilled water (S.D.W.) in sterile Petri dishes. *Pseudomonas* was then streaked onto King's medium B and *Xanthomonas* onto Yeast Dextrose Agar (Y.D.A.). Plates were incubated for 48 hours at 30°C. Single colonies were then restreaked onto the same media for purification.

PRINCIPAL MORPHOLOGICAL, PHYSIOLOGICAL, BIOCHEMICAL AND SEROLOGICAL CHARACTERISTICS

Xanthomonas campestris pv. **dieffenbachiae**

These characteristics were determined from 35 pathogenic bacterial isolates from anthurium collected from various locations in Guadeloupe and Martinique. These isolates were Gram negative, rod shaped with single polar flagellum.

The colonies were visible after 24 to 48 hours of incubation on LPGA and KY media. They were circular, yellow in colour and viscid. The isolates do not produce fluorescent pigments on King B medium. They are aerobic in type, and have an oxydative metabolism utilizing glucose. Cytochrome C oxidase was not detected nor was nitrate reductase. All the isolates demonstrated a hypersensitive reaction to tobacco.

Serological diagnosis: In the French West Indies, two isolates responsible for decline in anthurium (X2G2, origin Guadeloupe and X2M8, origin Martinique) and one isolate of **Xanthomonas campestris** pv. **dieffenbachiae** from Venezuela were selected for the production of immunoserums. These immunoserums were obtained from six months old rabbits, mixed with equal parts of glycerol and maintained at -20°C. We have used the indirect immunofluorescence method to determine the titre and specificity of the immunoserums. The 35 isolates had a positive reaction with the 3 immunoserums. The same reaction was observed with reference samples of **Xanthomonas campestris** pv. **dieffenbachiae**. The study of serological properties with respect to the 3 immunoserum prepared does not permit to differentiate the various bacterial isolates from Guadeloupe, Martinique, Venezuela and Hawaii. In addition, the serological reaction of isolates of anthurium is readily different from that of other members of the Araceae family. In the course of our study, we have observed that the isolates of anthurium can be easily distinguished from other pathovars of **X. campestris**. Only one crossed-reaction was observed with **Xanthomonas campestris** pv. **oryzicola**. The serological test allows to rapidly identify the **Xanthomonas campestris** pv. **dieffenbachiae** pathogen of anthurium.

Pseudomonas sp.

All isolates displayed similar characteristics which most closely associate them with the genus **Pseudomonas**. The bacterium rod-shaped was motile, Gram-negative, with a single polar flagellum. Colonies were circular, raised with an entire margin, and creamy white on KY, KA, KB and YDA medium. No fluorescent pigment was produced on KA or Kb. Some isolates produced a brown diffusible pigment when grown at 41°C on solid KY medium. No significant visible turbidity was determined after 10 days at 4°C or 41°C in liquid KY medium. All isolates elicited hypersensitivity in Xanthi and Samsun N.N. tobacco.

Serological Diagnosis: All isolates reacted positively when stained by the indirect fluorescent antibody technique. Antisera were produced from two representative isolates (PIG6, P4M9) from French West Indies. Anti-PIG6 and anti-P4M9 cross-reacted with **Pseudomonas rubrilineans**, and **Pseudomonas tolaasii** stained slightly when using anti-P4M9 sera.

Ps. rubrilineans and **Ps. tolaasii** were not pathogenic on anthurium when infiltrated into leaves. This serological test is useful for the quick and accurate identification of **Pseudomonas** pathogen of anthurium.

PATHOGENICITY TESTS

Plants inoculated with both pathogenic bacteria displayed symptoms identical to those observed under natural conditions. Symptoms on spray-inoculated anthurium usually appeared within 5-7 days under condition of high humidity. Soil-borne inoculations were positive resulting in vascular colonization. Bacteria interferes with the translocation of water and produce the development of characteristics symptoms.

CONTROL MEASURES

Resistant anthurium cultivars

It will be the most effective control measure but no variety appeared resistant as yet. Nevertheless, differences may be observed in varietal susceptibility of hybrid **Anthurium** spp. Under natural conditions, only hybrid **Anthurium** spp. seem susceptible to the **Pseudomonas** pathogen.

Sanitation

Presently, strict sanitation is the best control strategy for management of those diseases. Sanitation measures are recommended both at the commercial anthurium shelter-houses and country level:

- Nursery must be detached from productive plots.
- The sanitary state of the field must be checked.
- If diseased plants are found sanitize the field, remove symptomatic material and destroy all infected plants by burning. These operations will limit epidemics progression by reducing the number of bacteria.
- Cutting tools and clothes spread microorganisms: sterilize all tools with desinfectant to minimize pathogen dissemination and start cultural operations in plots with the lowest disease incidence.
- Water (splashing rain, irrigation) is accountable for rapid move of the pathogen: if possible sanitize during dry conditions and cultivate using aerated substratum.
- Transplanting plants from one place to another increases the possibility of introducing bacterial pathogens into uncontaminated areas: to avoid spread of disease restrict exchanges.

CHEMICAL CONTROL

Attempts to control these diseases with bactericides have been ineffective, and copper compounds have been reported to be phytotoxic to anthurium. Antibiotics have only bacteriostatic effects and will not stop multiplication of bacteria. Considering that it still exists a possibility of selecting antibiotic-resistant bacteria (uncontrollable), antibiotics should

not be used alone. Sanitation, by reducing inoculum level in the field, will minimize this possibility. Antibiotics (streptomycin sulfate and oxy-tetracycline) provide good control against bacterial blight only if used in conjunction with strict sanitation.

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