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HOSTPLANTS OF *AGROBACTERIUM GYPSOPHILAE*[1]

Waardplanten van Agrobacterium gypsophilae

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Symptoms obtained after either natural or artificial infection on roses, carnations and cacti by *Agrobacterium gypsophilae* are described, as well as a modification of the isolation method for this pathogen.

INTRODUCTION

Slowly increasing numbers of stunted roses, usually arranged about primary foci, have been noted in glasshouse crops at Aalsmeer. When these affected plants were examined, galls were found to be developing at the below ground level sites of grafting. Although the galls were reminiscent of those caused by *Agrobacterium tumefaciens* (Sm. & Towns.) Conn. attempts to isolate this pathogen, using a range of techniques and media, were unsuccessful and instead *Agrobacterium gypsophilae* (Brown) Starr & Weiss. was obtained.

METHOD OF ISOLATION AND PHYSIOLOGICAL PROPERTIES

White tumor-tissue was pulped in sterile saline solution (0.65% NaCl) which was then kept for 24 hours at room temperature before being streaked over nutrient agar. Nearly pure cultures of light yellow colonies developed after two days incubation. These cultures were not obtained if tissue was pulped in distilled water or peptone water, nor if the suspension was prepared less than ca. 24 hours before being streaked. Microscopically, the surface of the circular colonies, which had entire margins, were radially lined.

A. gypsophilae is Gram-negative, encapsulated and motile with 1–4 peritrichous flagellae (Fig. 1). Broth became turbid with a pellicle; growth in 5% NaCl was doubtful. Nitrites were not produced from nitrates. Hydrogen sulphide was produced. Methyl red and Voges-Proskauer tests were both negative. There was little or no liquefaction of gelatine. Litmus milk was peptonised and litmus reduced. Abundant growth occurred in Uschinsky and Cohn, but less in Fermi; pellicles were formed in all three but without fluorescence. Starch was not hydrolysed, and there was no lipolytic reaction. Acid was formed but no gas from glucose, sucrose, maltose, manitol and salicine. Acid was not produced in lactose. Carboxymethyl-cellulose was liquified, but pectin was not.

SYMPTOM EXPRESSION

Glasshouse roses

Spongy galls resembling clusters of tumorous adventitious roots, not consisting entirely of callus, developed only when grafts were buried below soil level (Figs. 2, 3). From them a few apparently normal roots developed, but at intervals along the latter smaller galls could be found. These symptoms appeared

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also six months after simultaneously inoculating and grafting. Subsequently the roots of the stock progressively died, the bush then depended on the normal adventitious roots of the graft. These symptoms on roses, caused by *A. gypsophilae*, are more or less intermediate between the tumors caused by *A. tumefaciens* and the hairy root symptoms caused by *A. rhizogenes*. Disease symptoms did not develop when stem bases of tobacco, *Pelargonium zonale*, and tomato were inoculated even though adventitious roots are formed by tomato as a normal response to wounding.

Glasshouse carnations

Because (1) roses and carnations are included in the crop rotation usually practised in glasshouses near Aalsmeer and (2) *A. gypsophilae* was first described from a member of the Caryophyllaceae, *Gypsophila paniculata*, the ability of this pathogen to attack carnations was tested. In some glasshouses plants were collected with large cortical cracks spreading from stem bases and in some instances with masses of tumorous callus growing from the pericycle (Fig. 4). *A. gypsophila* was isolated, not only from the tumors, but also from cracks on exposed surfaces and from necrotic areas high on the stem. As on roses, galls only occurred at the base of carnation stems where adventitious roots usually develop. These symptoms on naturally infected plants were reproduced by artificially inoculating the base of the stems. In addition round chlorotic spots developed on lower leaves, but *A. gypsophilae* could not be re-isolated from them (Fig. 5). Similar leafspots were found also on naturally infected plants.

C. Cacti

The cause of small tumors on cacti, at one time attributed to *A. tumefaciens*, was re-investigated and in some instances *A. gypsophilae* was isolated from galls formed of clusters of either spines or areoles (Fig. 6).

SAMENVATTING

Reeds enige jaren komen in kasrozen te Aalsmeer op sommige planten tumoren op de entplaats onder de grond voor. Hoewel dit verschijnsel werd toegeschreven aan een aantasting door *Agrobacterium tumefaciens*, werd dit pathogeen er nooit uit geïsoleerd. Door gebruik van een enigszins afwijkende isolatie-techniek werd daarentegen *Agrobacterium gypsophilae* geïsoleerd (fig. 1). De tumoren zijn sponsachtig en doen denken aan opgezwollen adventiefwortels (fig. 2, 3).

Eveneens werd de bacterie geïsoleerd uit de stengelvoet van kasanjers, waar de schors diepe scheuren vertoonde en soms een zuiver callusweefsel naar buiten kwam (fig. 4). Kunstmatig geïnfecteerde planten vertoonden daarnaast een necrose op de stengel en chlorotische bladvlekken (fig. 5).

Ook bij diverse cactus-soorten werden tumoren aan de planten gevonden, waar de bacterie in aangetoond kon worden (fig. 6).

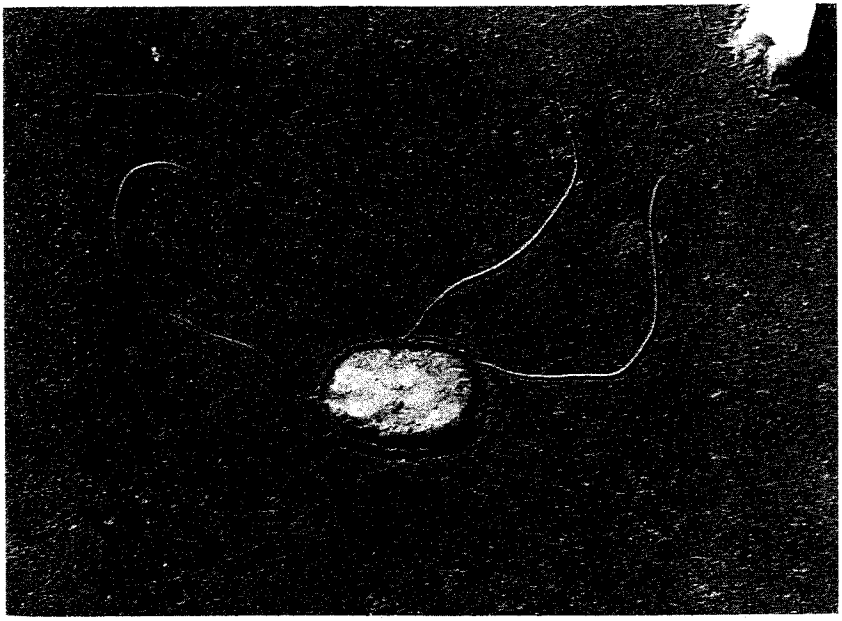

FIG. 1. *Agrobacterium gypsophilae* (Brown) Starr & Weiss. × 3000.

Photograph made by Physico-Technical Service for Agriculture, Wageningen.
Foto gemaakt door de Stichting Technische en Fysiologische Dienst voor de Landbouw Wageningen.

FIG. 2. Spongy gall on rose caused by *A. gypsophilae*.

Sponsachtige tumor op roos, veroorzaakt door A. gypsophilae.

FIG. 3. Radial section of a rose with a spongy gall. Grafting point clearly visible.

Radiale doorsnede van een rozestam met een tumor. De entplaats is duidelijk zichtbaar.

FIG. 4. Extremely large gall on the base of a carnation stem caused by *A. gypsophilae*.

Buitengewoon grote tumor op de basis van een anjerstengel, veroorzaakt door A. gypsophilae.

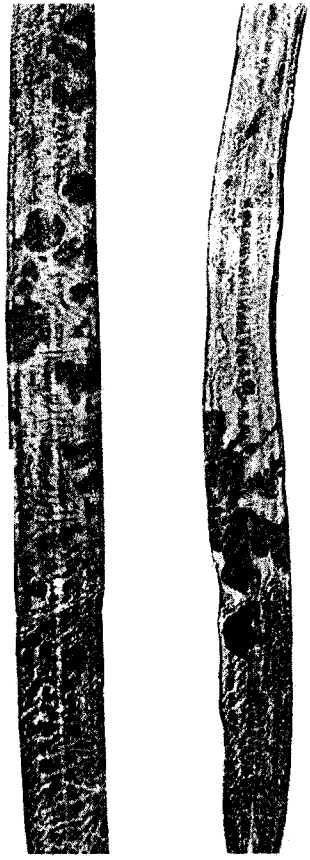

FIG. 5. Leafspots on carnetion infected by *A. gypsophilae*.

Bladvlekken op anjer, geïnfecteerd met A. gypsophilae.

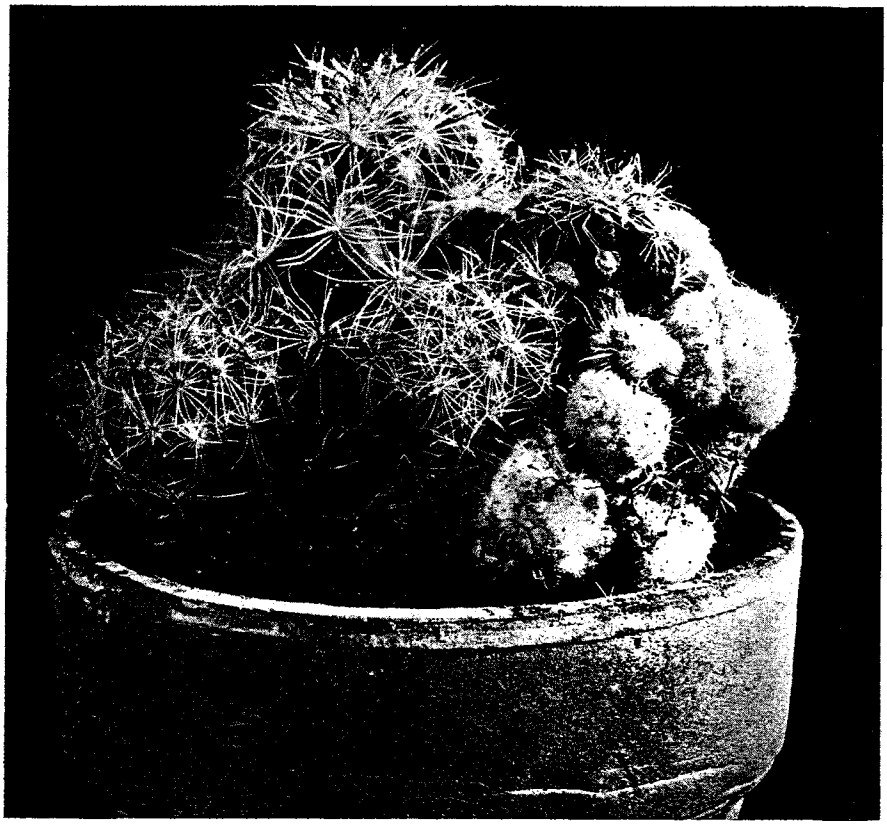

FIG. 6. Tumorous growth of *Mammillaria* spec. infected by *A. gypsophilae.*

Groeiafwijkingen van Mammillaria *spec., geïnfecteerd met* A. gypsophilae.