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Uncommon disease symptoms caused by Fusarium oxysporum in tulips forced in the glasshouse after pre-cooling at 5 °C

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Abstract

Rapidly developing disease symptoms not normally found in tulip bulb cultivation in The Netherlands are described. Mycelium of the fungus is present mainly in the parenchymatic tissues, giving further evidence that the fungus is not a vascular parasite in tulips. There are indications that enzymes or other compounds are involved in the syndrome. Possible factors influencing this uncommon disease development are mentioned.

Introduction

Fusarium oxysporum Schlecht. f. *tulipae* Apt does not behave as a vascular parasite in tulips under conditions prevailing during bulb cultivation in The Netherlands (Bergman, 1965). After planting in autumn, the plants growing from slightly infected bulbs or from healthy bulbs in contaminated soil, usually do not show any symptoms of the disease in the field. However, at lifting time (July) or during subsequent storage, the new bulbs grown from such plants often appear to be infected, thus demonstrating the presence of the parasite during the whole growth period. It has been shown that under these conditions the fungus often penetrates the side of the bulb and only occasionally infects the roots and the basal plate (Bergman, 1965). Premature death of tulip plants was observed only under experimental conditions of high temperature and inoculum potential.
The present paper describes a deviating disease syndrome observed during forcing of pre-cooled tulip bulbs in the glasshouse.

Deviational development of the disease

When tulip bulbs are cooled at 5 °C for 9–12 weeks prior to planting in a glasshouse in autumn or winter, the development of the plants is strongly accelerated. Whereas in normal culture 5–6 months elapse between planting and flowering, these cooled bulbs produce flowers in 5–8 weeks, depending on the pre-planting treatment and the soil temperature after planting (Hoogeterp, 1967).
Under these conditions, which have recently been adopted for flower production in winter, attack by *F. oxysporum* is very rapid. Growth retardation and yellowing of the leaves caused by this attack can be seen within 2 or 3 weeks after planting, often resulting in death of the plant before flowering. In such plants the fungus causes a decay

manifested as a greyish colour in the roots and the basal plate of the planted bulb. This decay spreads into the bulb scales and the lower part of the plant stem. Usually, fine dark longitudinal streaks are visible in the stem, suggesting a vascular disorder (Fig. 1).

Microscopical observations

Microtome sections of diseased tissues made after fixation in glutaric dialdehyde, dehydration in T.B.A., and embedding in diglycol stearate, were stained with saffranine-fast green or toluidine blue.
It is evident from these preparations that the fungus readily invades the parenchymatic tissue of the basal plate around the root bases (Fig. 2). At first it grows intercellularly, causing a dissolution of the middle lamellae and collapse of the cell contents (Fig. 3). In a later stage hyphae enter the cell lumina. In the adjoining vascular elements of the basal plate the fungus was found only occasionally. In a few instances root tips show invasion, the fungus growing upward through the cortex to the apparently undamaged root base. Usually, however, the root was colonized from the basal plate downward, hyphae growing abundantly both inter- and intracellularly in the cortex. Only when this tissue had been overgrown completely the fungus was found entering the vascular bundles.
In plants showing a discoloration of the stem base often accompanied by a yellowing of the leaves, mycelium was found in a relatively small part of the basal plate only and

Fig. 1. Tulip bulbs attacked by *Fusarium oxysporum* f. *tulipae* pre-cooled at 5 °C and planted at 16 °C soil temperature, a few weeks after planting. Discoloration and rotting of the basal plate, extending into the scales and the plant stem.

Fig. 1. Door Fusarium oxysporum f. tulipae aangetaste tulpebollen na koeling bij 5 °C geplant bij 16 °C bodemtemperatuur, enige weken na het planten. Verkleuring en verrotting van de bolschijf, uitstralend in de rokken en de stengel.

Fig. 2. Colonization by *F. oxysporum* f. *tulipae* of parenchyma surrounding the root base. Fungus has invaded the root cortex (below) radially, growing both inter- and intracellularly.

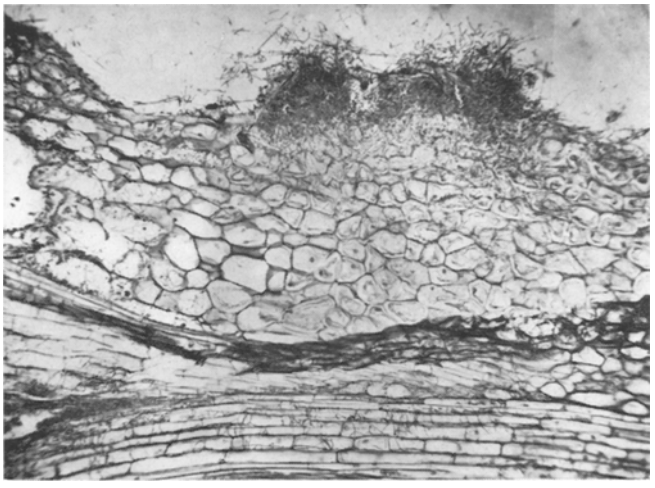

Fig. 2. Aantasting door F. oxysporum f. tulipae van het parenchym rondom de wortelbasis. De schimmel is doorgedrongen in de schors van de wortel (onder) en groeit daar zowel inter- als intracellulair in radiale richting.

Fig. 3. Tissue of the basal plate of a tulip bulb; fungal growth (*F. oxysporum* f. *tulipae*) mainly in the intercellular cavities of the parenchyma, which are often enlarged by dissolution of the middle lamellae. No hyphae visible in the vascular bundle.

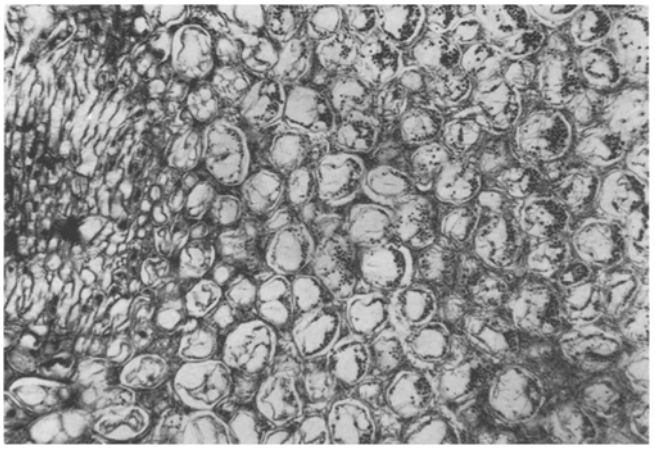

Fig. 3. Bolschijf van een tulp; de schimmel (F. oxysporum f. tulipae) groeit voornamelijk in de intercellulairen van het parenchym, die dikwijls vergroot zijn door oplossing van de middenlamellen. Geen hyfen in de vaatbundel.

Fig. 4. Transverse section through the lower part of a tulip plant stem with slight discoloration associated with infection by *F. oxysporum* f. *tulipae* of the basal plate. Vascular browning and clogging of xylem vessels; dissolution of phloem cell walls.

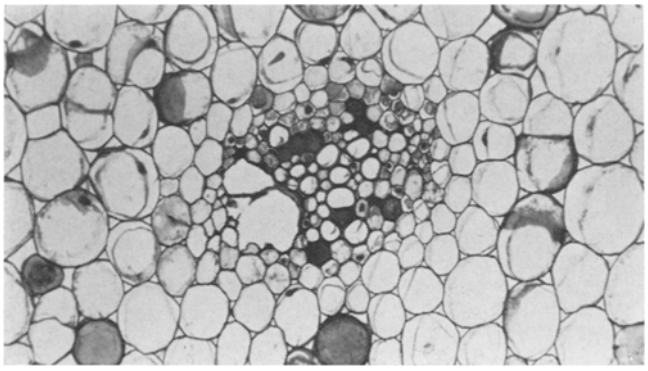

Fig. 4. Dwarsdoorsnede door het onderste deel van een tulpestengel, gegroeid uit een bol waarvan de schijf licht geïnfecteerd is met F. oxysporum f. tulipae. Verkleuring van wanden en verstopping van lumina van houtvaten; oplossing van celwanden in het phloeem.

not in the stem tissue itself. The walls of the xylem vessels in the stem are often slightly thickened and yellowish brown, and the lumina are clogged by a brown substance. Moreover, such vascular bundles show a collapse of phloem cells and a dissolution of cell walls of this tissue not observed in healthy plant stems (Fig. 4).

Discussion

The observations described here strongly support the conclusion that *F. oxysporum* is not a vascular parasite in tulips. Nonetheless, the consequences of infection are in some respects similar to those caused by vascular parasites of this genus in other host plants. Vascular browning and the formation of occlusions in the xylem vessels have been mentioned frequently in the literature (e.g. Waggoner and Dimond, 1955), as well as the discoloration of the parenchymatic tissue at a considerable distance from the loci at which mycelium was found. Probably, excretions by the fungus or metabolic products formed in the host–pathogen interaction are involved in the tulip strain in a way similar to that observed for other *Fusaria*. The presence of pectolytic and other enzymes has been demonstrated in *Fusarium*-diseased plants several times (e.g. Winstead and Walker, 1954). Since the fungus has been found growing in strongly enlarged intercellular spaces in the basal plate of tulip bulbs, the action of such enzymes in tulips seems probable. Maceration of phloem tissue, however, has been mentioned in literature only occasionally (Linford, 1931; Littlefield and Wilcoxson, 1962).
Although it seems evident that disorganization of stem tissue is induced long before there is mycelium growth, it must be emphasized that in several cases pieces of such stem tissue yielded *F. oxysporum* when plated on potato dextrose agar. It is therefore possible that conidia or mycelium fragments translocated through the vessels had escaped observation. It is not yet known why the attack by *F. oxysporum* is so rapid in

plants grown from bulbs pre-cooled at 5 °C, while this disease syndrome is rare in flowerbulb cultivation in The Netherlands. However, it is not uncommon in the cultivation in Japan (Schenk, 1967) and other countries. The soil temperature is certainly important. When grown in the glasshouse at 16 °C the plants usually die before flowering, while at 12 °C a marketable flower will be formed, though the bulb and the roots may show some symptoms of attack by *F. oxysporum*. Because of the temperature tolerance of the fungus, it seems improbable that a slight rise in temperature increases its virulence to such an extent. A higher soil temperature also accelerates root and shoot development considerably. It is supposed that both the prolonged cooling at low temperature and the very rapid outgrowth of the plant alter the metabolism of the tulip in such a way that attack by *F. oxysporum* is facilitated. Further work to investigate this hypothesis is in progress.

Samenvatting

Ongewone ziektesymptomen veroorzaakt door Fusarium oxysporum bij tulpen, welke in de kas tot bloei worden gebracht na koeling bij 5 °C voor het planten

Een snelle ontwikkeling van ziektesymptomen wordt beschreven, die bij de normale teelt van tulpen in Nederland niet bekend is. Mycelium werd hoofdzakelijk aangetroffen in de parenchymatische weefsels, zowel inter- als intracellulair. Dit geeft een verdere steun aan de opvatting dat de schimmel in de tulp geen vaatparasiet is. Er bestaan aanwijzingen, dat enzymen of andere stoffen een rol spelen bij het ontstaan van dit ziektebeeld. Faktoren, die wellicht een rol spelen worden genoemd.

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