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Neth. J. Pl. Path. 81 (1975) 154–156 SHORT COMMUNICATION

A device for the incubation of Fusarium-inoculated tulip bulbs in a constant air stream

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Inoculation experiments in tulip bulbs with *Fusarium oxysporum* Schlecht f. sp. *tulipae* Apt gave frequently inconsistent results when performed in stagnant air, even under strictly standardized conditions. This variability was thought to be due to the accumulation of ethylene produced in large quantities by the inoculum and by infected bulbs (Kamerbeek and de Munk, 1968; de Munk, 1972). Ethylene influences the metabolic processes in tulips, e.g. demonstrated by the induction of gum formation (Kamerbeek et al., 1971) and by the prevention of the de novo synthesis of tuliposid A in the outer scale after lifting (Beijersbergen and Bergman, 1973). The last phenomenon is of special interest, since on the presence of this compound the concept is based of a defence mechanism of the bulb against infection by *F. oxysporum* (Bergman and Beijersbergen, 1971).

The fungus grows well under anaerobic conditions, but was proved to produce hardly any ethylene (Kamerbeek and Swart, in prep.). Preliminary experiments showed that incubation of inoculated bulbs in an atmosphere without oxygen resulted in a considerable reduction of infections, while addition of ethylene had a reverse effect. Since bulbs and inoculum consume oxygen, it was probable that fluctuations in oxygen pressure may add to the inconsistency of results of experiments in stagnant air.

It was therefore felt necessary to avoid these disturbing influences in inoculation experiments meant to judge variations in susceptibility of bulbs during the growth period and storage, or to compare the susceptibility of various cultivars. For this reason a device was made and tested (Fig. 1) to incubate bulbs in a reproducably constant and

Fig. 1. Outline of a device for incubation of *Fusarium*-inoculated tulip bulbs in a constant moist gas flow.
A = pressure tank, B = dust filter, C = pressure regulator, D = air flow divider, E = capillary tubes, F = water bath, G = evaporator, H = condensor, I = incubation vessel.

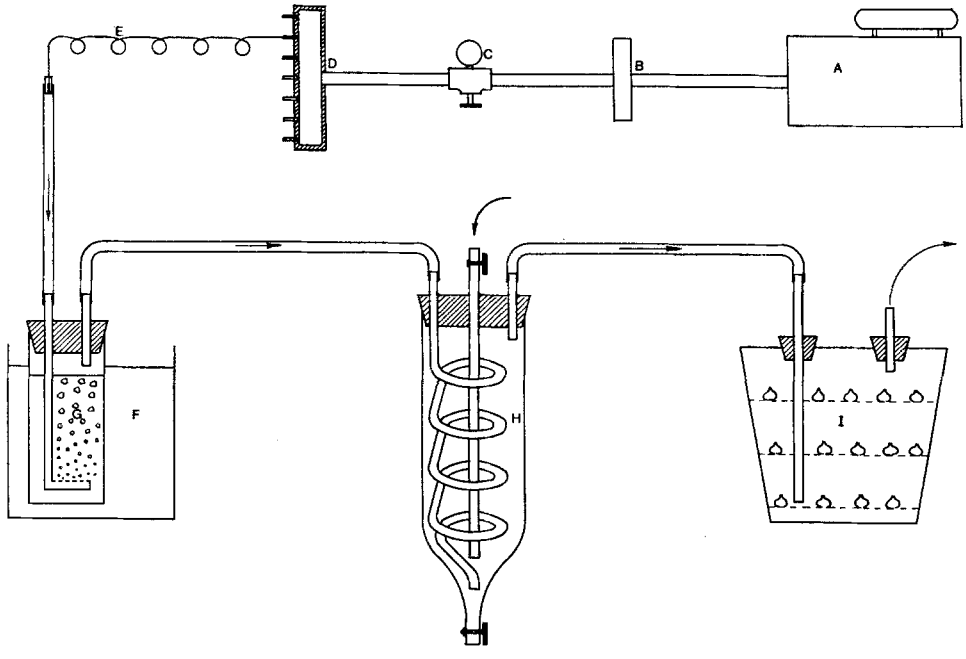

Fig. 1. Schema van een proef-opstelling voor incubatie, in een constante, vochtige gasstroom, van tulpebollen die geïnoculeerd zijn met Fusarium oxysporum.
A = drukvat, B = stoffilter, C = reduceerventielen, D = verdeelkop, E = capillairen, F = waterbad, G = verdamper, H = condensatievat, I = incubatie-ruimte.

water-saturated air stream, strong enough to keep the ethylene concentration at an inactive level (less than 0.05 µl/l; Kamerbeek and de Munk, in prep.), and without appreciable variations in oxygen pressure.

A 20-litre pressure tank (A) is fed by an oil-free compressor. After passing a dust filter (B), the air pressure is reduced by a two-stage pressure regulator (C). An air flow divider (D) is provided with a number of capillary tubes (E) of non-corrosive metal (inner diam. 0.5 mm). When each tube has the same length, the air speed at the outlets is equal. The flow rate is controlled such that at each outlet 24 litres of air per hour is released, viz. four times the volume of the incubation vessel (I). Each outlet is connected by plastic tubing (diam. 5 mm) with an evaporator (G) (vol. 0.5 litre) suspended in a waterbath (F) heated at 48 °C. The apparatus is placed in a room at 25 °C. The warmed and moistened air is led into a condensor (H) (vol. 1 litre), in which the air attains room temperature and becomes water saturated, after which it flows into the incubation vessel (I). Wet and dry thermometer readings in (H) and (I) do not vary, and small fluctuations of room temperature do not influence the internal conditions. Samples of air taken from vessel (I) during the incubation of 50 inoculated bulbs usually contained no more than 0.01 µl/l ethylene, except when many bulbs were heavily infected and 0.05 to 0.07 µl/l were found.

The third tube inserted in the stopper of (H) can be connected to a gas cylinder containing, for instance, ethylene, to be mixed with the moist air at a desired dose during circulation in the condensor. This does not influence the temperature or humidity of the gas flow at the outlet of (H), provided that the volume of the additional dry gas flow is less than 1/10th of the air flow.

The air in the system can be replaced completely by another gas by connecting the gas cylinder with the inlet of (D), or with one or more evaporators (G), using needle valves and air-flow meters to obtain the desired flow rate.

Koller and Thorne (1974) designed an apparatus to provide environmental conditions for plant physiological studies that is similar in several respects. The device described here permits equal distribution of constant air or gas streams to a large number of incubation vessels. The incubation conditions can be standardized for all vessels and the addition or substitution by other gases can be easily done without disturbing the conditions.

Samenvatting

Een proefopstelling voor de incubatie in een constante luchtstroom van tulpebollen, die geïnoculeerd zijn met Fusarium oxysporum

Inoculaties van tulpebollen met *F. oxysporum* f. sp. *tulipae* in stilstaande vochtige lucht gaven dikwijls niet-reproduceerbare resultaten.

Om de invloed te elimineren van variaties in ethyleen- en zuurstofconcentraties op het infectieproces, werd een opstelling ontworpen (Fig. 1), waarmee constante en met waterdamp verzadigde luchtstromen van de gewenste samenstelling en geschikte temperatuur gelijktijdig door een aantal incubatieruimten kunnen worden gevoerd. Dit wordt bereikt door samengeperste lucht middels een aantal capillairen in stromen van gelijke sterkte te verdelen, deze onder opwarming te bevochtigen en vervolgens door afkoeling tot omgevingstemperatuur met water te verzadigen.

De toevoeging van een ander gas of de vervanging van de lucht door een ander gas(mengsel) kan op eenvoudige wijze worden gerealiseerd.

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