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Extended Summaries SCI Pesticides Group Symposium Biological Control: Use of Living Organisms in the Management of Invertebrate Pests, Pathogens and Weeds

The following are extended summaries based on material presented at a meeting of the SCI Pesticides Group, held on 19–20 October 1992 at the SCI, 14/15 Belgrave Square, London SW1X 8PS, UK. The summaries published here are entirely the responsibility of the authors and do not necessarily reflect the views of the Editorial Board of Pesticide Science.

Bacillus subtilis—An Effective Biocontrol Agent

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Several strains of *Bacillus subtilis* (Cohn) Praz. have considerable antagonistic activity against soil-borne and foliar phytopathogens.[1,2] Although the complex mechanisms of their action are not fully understood, their use for biological control of plant diseases is under investigation.[3] We have isolated a series of strains from different sources and with a broad spectrum of antifungal and antibacterial activities during the last few years, and formulations containing spores of selected strains of *B. subtilis* have been developed in order to test their performance under practical conditions. Results of field and pot trials indicated differing contributions to disease control with selected host–parasite combinations, examples being delay of attack by and/or prevention of spreading of pathogens and effects on host plant growth.

A grower trial was performed over the period April 1986 to June 1987 in which the incidence of soil-borne carnation wilt, *Fusarium oxysporum* Schlecht f.sp. *dianthi*, (Prill. & Dell.) Snyder & Hansen on carnations in plots receiving different treatments (450 m^2 per variant) was compared. These treatments were the local standard treatment (sequential treatment with different fungicides), treatment with *B. subtilis* alone or with a combination of *B. subtilis* and zineb 800 g kg^{-1} WP (Agrokertrade, Budapest). The results, shown in Fig. 1, indicated that there were fewer diseased plants and a greater yield with the combined programme. Pot trials with tomato plants showed that treatment of the soil with *B. subtilis* resulted in improved root development with slips and increased yield of fruit.[4]

To study the conditions necessary for the establishment of *B. subtilis*, and to understand the main factors that influence the population dynamics of this antagonist in the soil, a strain marked by chromosomal antibiotic resistance was used. These processes were found to be dependent on the conditions during propagation and growth of the bacterial cells, the applied dose and the frequency of dosing, the time of application of the bacteria in relation to plant development stage, interactions with fungicides applied simultaneously (Figs 2 and 3) and the crop regime.[5] Moreover, the ability of the *B. subtilis* cells to produce significant amounts of antimicrobial metabolites seemed to be important for establishment and persistence.

Using several purification methods, especially chromatography, various antifungal substances were identified in supernatants from *B. subtilis* cultures. Analysis of the amino-acid content and [^{1}H]NMR/MS analysis of highly purified, biologically active fractions showed that the main antifungal substances were cyclic lipopeptides.[6,7]

Figure 4 shows the HPLC trace of material isolated

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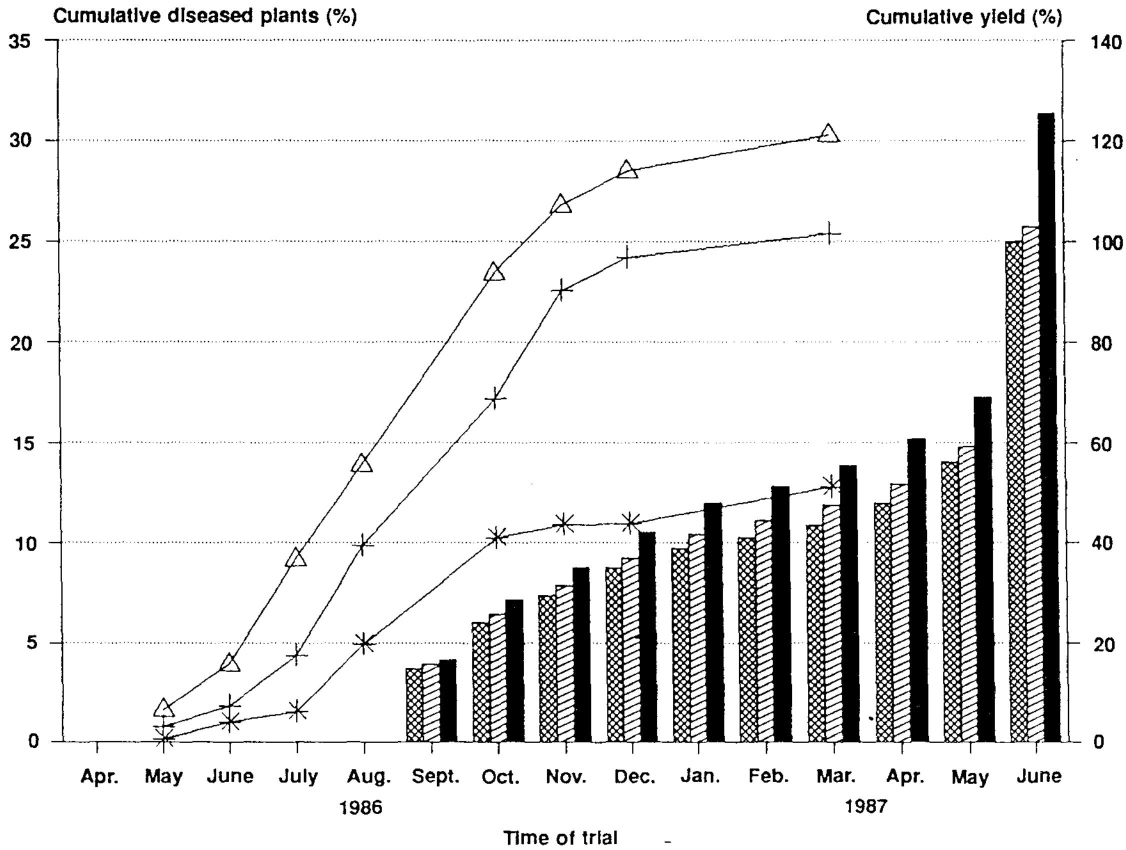

Fig. 1. Effect of *Bacillus subtilis* (+, ▨) alone or (*, ■) in combination with zineb on the incidence of wilt in glasshouse carnations, compared with (△, ⊠) the standard treatment, as indicated by the cumulative number of diseased plants and cumulative yields of flowers.

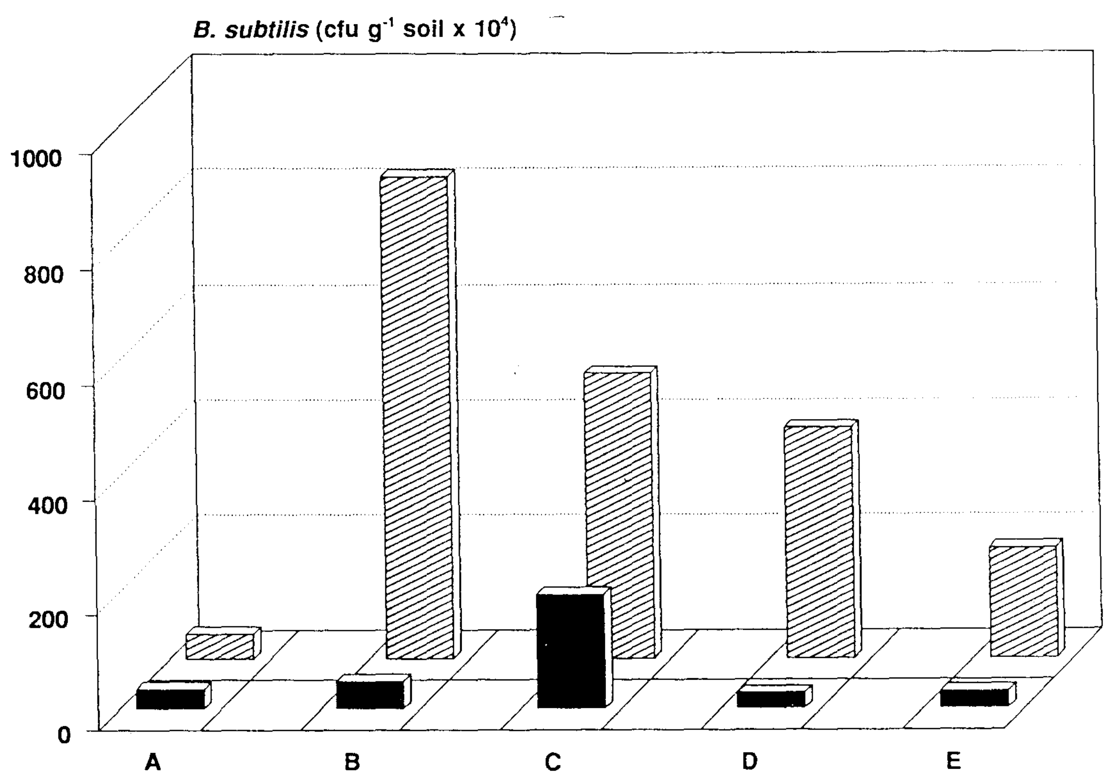

Fig. 2. Establishment of *Bacillus subtilis* (■) in soil and (▨) in the rhizosphere of pot-grown carnation plants. *B. subtilis* was applied (A) monthly, over four months (40 ml m^{-2}); (B) at the beginning of the trial (400 ml m^{-2}); (C) one month after the beginning of the trial (400 ml m^{-2}); (D) as in A but +zineb 800 g kg^{-1} WP (2 g a.i. litre^{-1}); (E) as in A but +zineb 800 g kg^{-1} WP (0·5 g a.i. litre^{-1}).

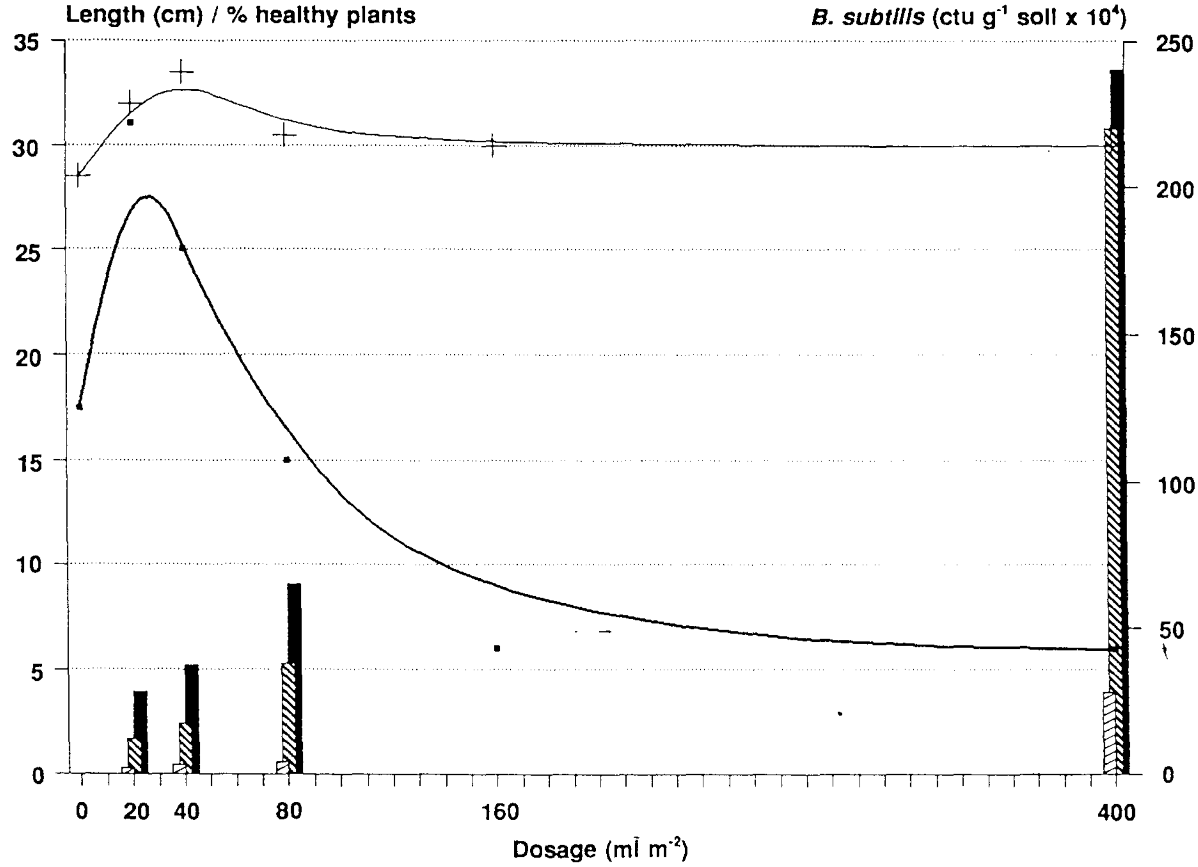

Fig. 3. Results of a pot trial (n = 10) with carnations to show the effects of *Bacillus subtilis* on carnation wilt, as indicated by the length (cm: +) and number (—) of healthy plants and the final population of *B. subtilis* (▨) 0–7, (▧) 7–14 and (■) 14–21 cm below the soil surface.

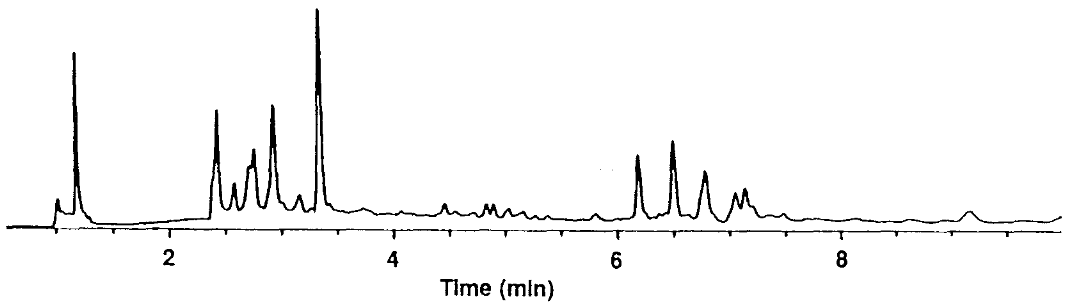

Fig. 4. HPLC profile of a supernatant from a culture of *Bacillus subtilis*. Peaks at retention times between 6·0 and 7·5 min mainly contain cyclic lipopeptides.

from the culture supernatant of a *B. subtilis* strain. This shows the presence of several cyclic lipopeptides eluting between 6·0 and 7·5 min and the similar elution times suggest only minor structural differences between the different compounds. For a given *B. subtilis* strain, the spectrum of compounds produced during fermentation differs little with time. However, both the actual and relative amount of a given peptide can vary according to the nutrients used in the culture medium.

Research is now being concentrated on the mechanisms of the antagonistic effects and monitoring of the establishment and persistence of the antagonist in order to improve the efficiency of *B. subtilis* for biological control.

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