

Biological control of some bacterial and fungal diseases on bean (*Phaseolus vulgaris* L.)

Fathy Gad Mohamed

SUMMARY The present work was carried out to study the role of the biological control of diseases caused by *Rhizoctonia solani*, *Sclerotium rolfsii* and *Corynebacterium flaccumfaciens* in bean cultivars. These studies have been done under laboratory, greenhouse and field conditions. Results obtained can be summarized as follows: 1- *Rhizoctonia solani*, *S. rolfsii* and *C. flaccumfaciens* isolates varied in their pathogenicity against different bean cultivars. 2- Different microorganisms used in these study as antagonists were isolated from sclerotia of pathogenic fungi, rhizosphere of bean plants and identified isolates obtained from Bacterial Dis. and Biological Control Dept. Plant Pathology Inst. Agric. Res. Center, Giza, Egypt and Fungus and Plant Pathology branch, Dept. Agric. Botany. Fac, of Agric. Moshtohor. 3- An aggressive isolates of *R. solani* and *S. rolfsii* were used under laboratory conditions, to study the effect of physiological factors affecting antagonism between pathogenic fungi isolates and different antagonistic fungi and bacteria. Reaction in mycelial growth or sclerotial formation of *S. rolfsii* were used as two parameter to measure the efficacy of the antagonist. 4- Data obtained under laboratory conditions show that, different antagonists varied in their reaction according to different physiological factors. Glucose, fructose and sucrose were the most favourable carbon source for the antagonistic effect of against *R. solani*. Dextrin was the least one effecting the antagonistic effect of fungi against *R. solani*. While, lactose, starch, dextrin were the most favourable carbon source for the antagonistic effect against *S. rolfsii*. Starch, sucrose, fructose and glucose were the most favourable carbon source for antagonistic bacteria against *R. solani*. On the other hand, lactose, glycerol and starch were the most effective carbon sources on the antagonism between antagonistic bacterial isolates and *S. rolfsii*. 5- Ammonium tartrate and potassium nitrate were the most favourable nitrogen sources for antagonism between different antagonistic fungi and *R. solani* and *S. rolfsii*. On the other hand, urea and potassium nitrate were the most favourable nitrogen sources for antagonism between different antagonistic bacteria and *R. solani*. Sodium nitrate, ammonium nitrate and potassium nitrate were the most favourable nitrogen sources for antagonistic effects of different antagonistic bacterial isolates against *S. rolfsii*. 6- CIN ratios 15/2 and 25/5 were the most suitable ratios for antagonistic effect of different antagonistic fungi and *R. solani*, whereas CIN ratio 00/2 and 25/2 were the most suitable ratios for antagonisms between different antagonistic fungi and *S. rolfsii*. On the other hand, CIN ratios 1/0.5, 5/2.5 and 2/1 were the most suitable ratios for antagonistic effect of different antagonistic bacterial isolates against mycelial growth of *R. solani*. While CIN ratio 3/1.5 was the most suitable ratio for antagonism against *S. rolfsii*. 7- Regarding the effect of pH value on the efficacy of different antagonists, it was clear that, at pH 6.5 and 7 were the most suitable pH values for the antagonistic effect between antagonistic fungi and pathogenic fungi (*R. solani* and *S. rolfsii*) causing increasing of reduction in mycelial growth and reduction in sclerotial formation of *S. rolfsii*. On the other hand, pH 6.6 and 7.6 were the most suitable pH value for the antagonistic effect of antagonistic bacterial isolates and *R. solani*. While pH 6.6 and 7.0 were the most pH value effects against mycelial growth of *S. rolfsii*. 8- Regarding the effect of temperature degrees on the antagonism, 25 and 30°C were the most suitable degrees for antagonists effect against mycelial growth of *R. solani*. While, 20 and 25°C were the most suitable degrees for antagonisms between different antagonistic fungi and *S. rolfsii*. On the other hand, 30°C were more effective than 25°C on the antagonism

between different antagonistic bacterial isolates and *R. solani* or *S. Rolfsii*. 9- Morphological studies on the effect of antagonistic bacteria *B. subtilis* on mycelium of *R. solani*. After 4 days showed that, mycelium of *R. solani* showed malformed and lytic. 10- Histological studies, showed that, *T. harzianum* parasitized sclerotia of *S. rolfsii* and absolutely destroyed it within 10 days under laboratory conditions. 11- Adding culture filtrates of different antagonistic fungi after 5, 10, 15 and 20 days to the medium of *R. solani* and *S. Rolfsii* led to reduction in mycelial growth of the pathogenic fungi. Data obtained revealed that, positive correlation between the percentage of reduction and rate of adding culture filtrate and age of culture filtrates. Culture filtrates of *T. harzianum* iso. No. 28 was the most toxic ones to *R. solani* mycelium while *T. harzianum* iso. No. 5 was the most effects against *S. Rolfsii*. On the other hand, slight differences between *T. harzianum* iso. No. 3, 4 and NO. 5 on reduction in amount of mycelial growth of *R. solani*. *T. harzianum* iso. No. 3 was the most toxic one against *S. rolfsii* to reduction in amount of mycelial growth was filtrates taken after 15 days of incubation. 12- Effect of time inoculation, of the antagonists, on the mycelial growth of *R. solani* and *S. rolfsii* or sclerotial formation of *S. rolfsii* were also tested. Data obtained revealed that, positive correlation between incubation period and percentages of reduction in mycelial growth of *R. solani* and *S. rolfsii* or sclerotial formation of *S. rolfsii* were recorded. All isolates of *T. harzianum* showed more antagonistic effect than *G. penicilloides* on reduction in mycelial growth and sclerotia formation of *S. rolfsii*. On the other hand, *Bacillus* iso. No. 2 and *B. cereus* were the most effect against *R. solani*, while *B. subtilis* iso. No. 2 and *Ps. fluorescens* were the most effects against *S. Rolfsii*. 13- Under greenhouse conditions inoculum potential of the aggressive pathogenic fungi and methods of inoculation for the pathogenic bacterial isolates indicated that, 6 g/kg of soil for *R. solani* and 75 sclerotia/kg of soil for *S. rolfsii* were enough to destroy highest in % of bean plants. While methods of inoculation of *C. flaccumfaciens* varied between different bean cultivars. 14- Under greenhouse conditions, *C. flaccumfaciens* as a causal organism responsible for wilt disease in bean plants, was able to infect soybean, pea, cowpea and water-melon. While broad bean, tennis, tomato, squash, rice and lettuce were not susceptible to *C. flaccumfaciens*. 15- Different antagonistic fungi were used as spore suspension or culture filtrates to control disease incidence caused by *R. solani* and *S. rolfsii* in bean compared with fungicide vitavax-captan 75% under greenhouse conditions. Data obtained revealed that, spore suspension was more effective on control disease incidence than culture filtrates or vitavax-captan and caused increase in % survival plants, fresh, dry and No. of pods/pot. On the other hand, *T. harzianum* iso. No. 28 were more effective than other isolates. 16- Under the same conditions, seed or soil treated with antagonistic bacterial isolates to control disease incidence caused by the same pathogenic fungi in bean cvs. Data obtained revealed that, in regard to Giza 3 cv soil treated with different antagonistic bacteria was more effective on reducing in % of disease incidence than seed treatment which caused increase in % of survival plants. While seed treated with vitavax-captan was more effective than bio-agents in Contender and Bronco cvs. On the other hand, all biological agents were more effective on increasing No. of pods/pot than fungicide in all bean cvs. Regarding fresh and dry weight/pot data indicate that, isolates of antagonistic bacteria varied in their effects according to reaction between the variety and the pathogens, and showed more or equal with chemical treatment in increasing fresh and dry weight/pot for bean cvs. 17- Regarding the effect date of application of biocontrol agents against pathogenic fungi on disease incidence caused by *R. solani* and *S. rolfsii* in bean cvs. under greenhouse conditions. Adding different antagonists before or at sowing were more effective on reduction of disease incidence than any other ways compared with fungicide treatment in case Giza 3 and Contender cvs. when used against *R. solani*. Regarding No. of pods/pot *B. subtilis* was the most antagonist effective in increasing No. of pods/pot when added to soil before sowing in Giza 3 cv. While *T. harzianum* and *Ps. fluorescens* increased No. of pods/pot when added at three times or at sowing respectively. Fresh and dry weight of bean plants was clear when different antagonists were added at three times. 18- Regarding the effect of application time of adding antagonistic bacterial isolates on % wilt disease incidence caused by isolates of *C. flaccumfaciens* in bean cvs. Data obtained revealed that, adding different antagonistic bacterial isolates at 72 hrs before inoculation of pathogenic isolates were more effective on reducing in % wilt disease incidence in bean cvs. than adding at the same time. On the other hand, Contender cv was the most affected by using antagonistic bacterial isolates by only 9.2%

wilt diseased. While Bronco and Giza 3 cvs were slightly affected by them and 30.3 and 42.50/0 wilt diseased were obtained respectively. *B. cereus* was the most antagonists effective against isolates of *C. flaccum/aciens* and caused only 22.5 % wilt disease. While no differences between *B. subtilis* iso.No.2 and *Ps. fluorescens* in controlling *C. flaccum/aciens* isolates in bean cvs. by caused 32.2 % wilt diseased.

19- Comparison between the effect of different antagonists as seed dresser or soil treatment on disease incidence caused by *R. solani* and *S. rolfsii* in bean cvs. Data obtained revealed that, adding different antagonists as soil treatment (granules on wheat straw) or as seed dresser were more effective than other forms on disease incidence in Giza 3. On the other hand, *T. harzianum* and *B. subtilis* were more effective than *Ps. fluorescens*. Seed treated with different antagonists increased in No. of pods/pot in Giza 3 cv. than other ways. Slightly differences between different treatments in increasing fresh and dry weight/pot.

20- Under field conditions, the same three antagonists, *T. harzianum*, *B. subtilis* and *Ps. fluorescens* were used as seed or soil treatment and different application dates for bean cvs during 1995 and 1996 seasons. Data obtained revealed that, *T. harzianum* and *B. subtilis* used as seed treating were the most effective on reduction in disease incidence and increased yield of bean cvs comparing with control or fungicide vitavax-captan.

21- Adding any of the tested antagonists two weeks before sowing seeds, inhibited disease incidence and increased the yield of bean cvs., more effectively if compared with adding at divided into two amounts or at two weeks after sowing.

22- Under the same conditions, adding different antagonists at different forms led to different degrees of protection against disease incidence in bean cvs. Also, different antagonists behaved differently according to the variety of treated plants. In general, using antagonistic fungi as granules (on wheat straw or bran) led to the most protection of disease incidence, and increased of yield bean cvs. Regarding, the antagonistic bacteria, no trend was observed among different forms in most cases for controlling of disease incidence and yield of bean cvs during 1995 and 1996 seasons.