

pathological studies on cotton seeds used in agriculture

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Cotton (*Gossypium hirsutum* L.) is one of the most important fiber and oil crops in Egypt and many other countries all over the world. It is attacked by several disorders, which resulted from insects, fungi, bacteria, nematodes and others at the different stages of growth. Cotton seedling diseases whether pre or post emergences are world wide problem, often causing serious stand losses. A number of soil and seed borne pathogens can infect cotton seedlings individually or in association as a disease complex. Cotton plants are subjected to attack by various pathogenic fungi causing several diseases during different stages of growth. . the obtained results of the present study could be summarized as follows : 1-Isolation trials from different parts of cotton seeds of cvs Giza-86 and Giza-89 (before and after delinting, testa and cotyledons) resulted in several fungi which belonging to 5 genus and 11 species. The isolated fungi were purified and identified as *Alternaria alternata*, *Aspergillus niger*, *Fusarium dimerum*, *Fusarium moniliforme*, *Fusarium nivale*, *Fusarium roseum*, *Fusarium semitectum*, *Fusarium tricinctum*, *Fusarium solani*, *Penicillin spp* and *Rhizoctonia solani*. As well as, some isolated fungi stayed unknown without identification, t-The isolated fungi from different parts of cotton seeds as well as rotten roots of Giza-86 and Giza-89 were differed in their frequencies from part to another. Generally, *R. solani* was the highest frequent fungus followed by *Fusarium* meanwhile, only the infected plants with *F. semitectum* revealed protein bands more than in control treatment.14-On the other hand, all infected cotton plants (Giza-89) produced few of new bands as in response to infection with root rot pathogens like 17.5 KDa in case of *R. solani*, 20.1 KDa with *F. moniliforme*, 82.9 and 21.6 KDa with *F. roseum* as well as, 130.9, 99.5, 96.3, 46.4, 36.3, 30.4 ,16.1 and 13.8 KDa in case of *F. semitectum*. On the other hand, the inoculated cotton plants (Giza-89) with *F. roseum* and *F. semitectum* revealed typical protein band at 21.6 KDa, 15-Infestation the soil with tested root rot pathogens i.e. *R. solani*, *F. moniliforme*, *F. semitectum*, and *F. roseum* before sowing cotton seeds in this soil do not affect esterase isozyme content in cotton leaves of growing seedlings at 21 days old in case of cvs Giza-86 and Giza-89 comparing to un-infested one (control). In this respect, all resulted protein patterns of esterase isozyme of all infestation cases were equal to those appeared with control treatment where they taken the same values of Rf. The only exception was for *R. solani* which formed one new protein band at 0.36 with cv Giza-86 and five new protein bands of esterase isozyme at Rf values 0.21, 0.36, 0.62, 0.71 and 0.79 with cv Giza-89, 16-Infestation the soil with tested root rot pathogens i.e., *R. solani*, *F. moniliforme*, *F. semitectum*, and *F. roseum* before sowing cotton seeds in this soil incited clear determined phenols content were higher in general in cv. Giza-89 than those determined in case of Giza-86.9-Infestation the soil with tested root rot pathogens i.e. *R. solani*, *F. moniliforme*, *F. semitectum*, and *F. roseum* before sowing cotton seeds in were affected negatively the content of chlorophyll when determined in leaves of cotton seedlings of both tested cvs after 21 days post sowing. In this respect, all the tested pathogens decreased the content of chlorophyll A and B and then total chlorophylls in cotton leaves comparing with uninfested soil (control) of both two cvs (Giza-86 and Giza-89). The highest decrease in chlorophyll A and B as well as total chlorophyll was recorded in case of infestation the soil with *F. moniliforme*, *F. semitectum*, *F. roseum* and *R. solani* respectively comparing with uninfested soil (control) of both two cultivars. The results indicated also that infestation the soil before sowing with root rot pathogens do not affect clearly on carotenoids

content in leaves of both tested cvs comparing with un-infested soil (control). 10-As for the variation in fractionated protein patterns of two cotton cultivars (Giza-86 and Giza-89) due to infection with *Rhizoctonia solani* and *Fusarium* spp, it is pronounced that the infection with those pathogens increased the number of fractionated protein bands comparing with control treatment (un-infested). In this respect, the infected cotton plants (Giza-86) with *R. solani* revealed 15 protein bands comparing with control plants (11 band), some of them are similar in their molecular weight to as in control while some, others are newly formed corresponding to infection with the pathogen, 11- Meanwhile, infection with *F. moniliforme* and *F. roseum* cv. Giza 86 revealed 12 protein bands, some of them are new formed. On the other hand, the inoculated cotton plants (Giza-86) with *F. semitectum* revealed 16 protein bands comparing with the un-inoculated ones (11 band) where the new formed bands were 192.6, 148.4, 116.2, 93.1, 47.7, 40.1, 28.9 and 16.1 KDa. The results indicate also that the inoculated cotton plants (Giza-86) with *F. roseum* and *F. semitectum* revealed typical protein bands at 192.6, 148.4, 93.1 and 16.1 KDa as well as they differed completely with those protein bands of *R. solani* infection, meanwhile they were similar partially with the inoculated with *F. moniliforme* at 16.1 KDa, 12. Moreover. The inoculated cotton plants (Giza-86) with *R. solani* and *F. moniliforme* produced typical bands in response to infection at 100.0, 40.1 and 17.5 KDa. These appeared protein bands are differed in their condense where some of them are clear faint and some others appeared intensive although they have the same molecular weight, 13- As for cotton cv Giza-89, there were clear variations in fractionated protein patterns in leaves due to infection with *Rhizoctonia solani* and *Fusarium* spp. These exhibited protein bands were ranged between 208.6-11.9 KDa. It is pronounced that the infection with *R. solani* and *F. roseum* revealed protein bands lesser in their number comparing with control treatment (un-infested) variations in peroxidase isozyme patterns of two tested cotton cvs Giza-86 and Giza-89, 17-As for Giza-86, it is pronounced from the obtained results that the infestation with *R. solani* formed two new protein bands at Rf 0.10 and 0.12 comparing with control treatment (un-infested), meanwhile., the infestation with *F. moniliforme* do not reveal any new bands comparing with control treatment (un-infested). whereas, the infestation with *F. semitectum*, and *F. roseum* revealed only one new band of peroxidase isozyme at Rf. 0.10. 18-Concerning cv Giza-89, infestation the soil with tested root rot pathogens resulted in clear changes in formation of peroxidase isozyme where many protein bands were disappeared comparing to those of un-infested one (control), meanwhile, no one of new protein bands were formed with all infestation treatments. 19-Infestation of cotton seeds with tested root rot pathogens i.e. *R. solani*, *F. moniliforme*, *F. semitectum*, and *F. roseum* decreased the percentages of oil content into both tested cotton seeds of cvs Giza-86 and Giza-89 comparing with uninfested seeds (control) at all incubation days which ranged between 5-15 days. It is clear also that increasing incubation days from 5-15 days decreased gradually the determined percentages of oil contents for all tested pathogens comparing with un-infested seeds (control). The highest decrease in percentages of determined oil contents were recorded in case of infestation the seeds with *R. solani* , and *F. moniliforme* at all tested incubation days for seeds of both cotton cvs. 20-Regarding mycotoxin production, of the tested fungi that isolated from cotton seeds were not able to produce any of aflatoxins (B 1 & B2), zearalenone, fumonisins and trichothecenes when grown in vitro on specific YES medium, 21-On the other hand, infestation the cotton seed samples of both cvs (Giza-86 and Giza-89) with those tested root rot pathogens produced clear amounts of mycotoxins (ppb) in some cases. In this respect, *F. semitectum* and *F. roseum* produced zearalenone mycotoxin onto infected seeds of cv. Giza-86 and cv. Giza-89 while, *R. solani* and *F. moniliforme* were not able to produce Zearalenone mycotoxin onto cotton seeds of both tested cvs. As for fumonisins mycotoxins, only *F. moniliforme* produced onto infected seeds of Giza-86 and cv. Giza-89. In addition, no one of the four tested isolates was able to produce aflatoxins onto infested cotton seeds, meanwhile it is pronounced that aflatoxins were appeared only on naturally contaminated cotton seeds of both tested cvs, 22-All of tested fungicides reduced the growth of all the tested pathogens. In this respect, they reduced the linear growth of *R. solani* where Premis, Maxim and Topsin-M were the best effective fungicides. It is clear also that Topsin-M was the highest effective one at the concentrations 5-400ppm followed by Premis and Maxim at concentrations of 10-400ppm. On the other hand. Maxim and Primes were more effective than

Topsin-M at concentration 1 ppm. Moreover, the effective fungicide in reducing the growth of *R. solani* was Vitavax-T70. Also increasing the concentration from 1 to 400ppm increased gradually the effect of tested fungicides in reducing the growth of *R. solani* where the concentration like 200 and 400 ppm were more effective than others.²³ Also, Maxim and Premis as well as Topsin-M were the , most effective fungicides in reducing the growth of *Fusarium moniliforme*. On the other hand, Rizolex-T, Vitavax-T70 and Vitavax-T40 were less effective than the first three fungicides although they reduced the growth of *F. moniliforme*. On the other hand, Premis followed by Topsin-M were the highest effective fungicides in reducing the growth of *Fusarium roseum* and *Fusarium semitectum* more than other tested fungicides. Premis fungicide and Topsin-M gave the highest reduction at all tested concentrations except 1 ppm for the second.²⁴ Concerning the effect of bioagents, Rizo-N and Plantguard bioagents reduced the linear growth of all tested root-rot pathogens comparing to un-treated one (control) where, Rizo-N and Plantguard succeeded in reducing the linear growth of *R. solani*, *F. roseum*, *F. moniliforme* and *F. semitectum* to remarkable values. Moreover, Rizo-N was better than Plantguard in its effect in reducing the linear growth of all tested fungi.²⁵ As for the pathogenic potentialities of tested fungi whether individual or in combination in presence or absence of any antagonists before sowing in normal soil without infestation (control-2) with any of tested root rot fungi improved the shoot length of resulted seedlings in case of Maxim, Vitavax-T40, Rizo-N and Plantguard treatments more than other treatments and control. In general, Rizolex-T and Maxim were the best tested fungicides in increasing the shoot lengths of resulted seedlings of cv Giza-86 while, Rizo-N, Topsin-M and Vitavax-T70 were the best in case of cv Giza-89.³² Also, treating cotton seeds cv Giza-86 with fungicides or antagonists before sowing in infested soil with tested pathogenic fungi reduced significantly the root length, and dry weight of resulted seedlings in most cases of treatments comparing with un-treated seeds in infested soil with tested fungi (control-1). However, some fungicides treatments increased the seedlings root length like Premis seed treatment with soil infested with *R. solani* and soil infested with *F. semitectum*. As well as, there was a clear increase in root length in case of Maxim and Vitavax-T70 and Plantguard seed treatments before sowing in soil infested with *F. semitectum*.³³ Also, treating cotton seeds with fungicides or antagonists before sowing in normal soil without infestation with any of pathogenic fungi (control-2) improved the root length of resulted seedlings more than un-treated seeds in normal soil (control-1). On the other hand, treating the cotton seeds cv Giza-89 with Topsin-M fungicide before sowing in infested soil with *R. solani* or *F. moniliforme* only increased the root length of resulted seedlings comparing with other fungicides or antagonists treatments. Meanwhile, all seed treatments with fungicides or antagonists before sowing in infested soil with *F. semitectum* decreased the root lengths of resulted seedlings to lengths lesser than that of control-1 treatment. On the other hand, seed treatment with Rizolex-T was the best treatment in case of infested soil *F. roseum* in increasing significantly the root length of resulted seedlings comparing with other treatments and control.³⁴ In the same time, treating the seeds of Giza-89 with fungicides or antagonists before sowing in normal soil (control-2) reduced the root lengths compared to control-1 treatment. Also, treating the cotton seeds cv Giza-89 with any of fungicides or antagonists before sowing in soil infested with *R. solani* reduced the dry weight of resulted seedlings at 21 days old. While all seed treatments with any of tested fungicides and antagonists before sowing in infested soil with *F. moniliforme* increased significantly the dry weight of resulted seedlings comparing with control-1 treatment where Vitavax-T70, Maxim, Premis and Topsin-M were the best seed treatments in this respect. Also, Rizo-N and Premis were the best seed treatments before sowing in soil infested with *F. semitectum* in increasing the dry weight of resulted seedlings comparing with control-1 treatment whereas other seed treatments reduced significantly the dry weight of resulted seedlings. On the other side, Rizo-N and Vitavax-T40 were the best seed treatments before sowing in the normal soil (control-2) in increasing the dry weight of resulted seedlings in un-infested soil comparing with control-1 (un-treated seeds).³⁵ On the other hand, treating cotton seeds with fungicides and commercial bioagents reduced root rot incidence under field conditions. Maxim and Topsin-M followed by Vitavax-T70 were the best effective fungicides on reducing root-rot incidence of tested cotton plants (cvs Giza-86 and Giza-89) during growing seasons 2000 and 2001. Also, Rizo-N was the best effective bioagent on reducing root-rot

incidence and increasing the survived plants of both cotton cvs during the two growing seasons.36- Meanwhile, treating cotton seeds with fungicides or commercial bioagents before sowing for controlling root rot pathogens increased also plant height, number of fruiting branches on growing plants. number of the opened bolls/plant. weight of yielded cotton for each boll, cotton yield (g)/plant, average yield of cotton lint (g)/plant. yield of cotton seeds (g)/plant. yield of cotton (kantar/feddan). yield of cotton lint (kantar/feddan). yield of cotton seeds (kg/feddan) and fiber length of yielded cotton of both cotton cvs (Giza-86 and Giza-89) during the two growing seasons comparing to un-treated seeds (control). Generally, Vitavax-T70, Vitavax-T40 and Rizo-N were the best seed treatments in this respect. It is pronounced also that Plantivard and Rizo-N are more effective than many of tested fungicides like Maxim, Topsin-M and Rizolex-T as well as un-treated ones (control).