

Efficacy of Bioagent in Controlling Soil Borne Fungi and Assessment of Seed Yield and its Components in Peanut (*Arachis Hypogea*)

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ABSTRACT

The study was carried out on the experiment field of Ismailia Agricultural Research Station, Ismailia Governorate during the two successive summer seasons (2019 and 2020) using two genotypes of peanut (Giza 6 & Intro. 293). To study effect of bio-agent such as *Streptomyces erythrogriseus* 2 (100%, 75% and 50% cell suspension) and fungicide (Rizolex T 50% WP) on controlling soil borne fungi (*Fusarium semilectum*, *Aspirigillus niger*, *Aspirigillus ochraceus*, *Tricoderma* sp., and *Penicilium* sp.) on peanut crop. Results indicated that Intro. 293 was surpassed cultivar Giza 6 in most of growth characters and seed yield parameters. The concentration of 50% bio-agent *Streptomyces erythrogriseus* 2 with the two studied genotypes gave the highest result in chlorophyll a (55.10). The highest seed oil percent (52.43%) for Intro. 293 was observed with concentration 75% of bio-agent *Streptomyces erythrogriseus* 2, in the second season. The growth of the *A. flavus* fungus, *Alternaria alternaria* and *Cladosporium* sp. was inhibited 100%, when, *Streptomyces erythrogriseus* sub sp 2 treatment through all concentrations compared to control, seeds were subjected to seed healthy test. The percentage was less in case of *Fusarium semilectum*, *Aspirigillus niger*, *Aspirigillus ochraceus*, *Tricoderma* sp., and *Penicilium* sp. Phenolic compounds test showed that concentration 100% filtrate give the heights result 20.38 µg/g of fresh weight. CO₂ test showed high percent in concentration 100% filtrate, 187 mg/100mg soil. An economic evaluation of the experiment was also conducted, where the concentration of 75% of bio-agent with the Intro. 293 gave the highest production value per acre of 9441.1322 pounds per acre in the first and second seasons.

Keywords: Actinobacteria, bio-agent, fungicides, Peanut, soil borne fungi, *Streptomyces*, and yield components.

INTRODUCTION

Peanut is an important oil and protein crop, which is grown mainly in semi-arid tropic and sub-tropic areas of countries around the world (Siva *et al.*, 2014). Peanut cultivated area in the world is approximately 34.9 million hectare and productivity of 72.6 million ton (FAO STAT, 2021). On the other hand, in Egypt peanut is cultivated in many Governorates such as Ismailia, El-

Sharkia, and El-Behera, where peanut crop ranked as the first among the cultivated oil crops in Ismailia Gov. with rate of seed yield production 13.2 thousand ton (BECAMS, 2023).

The peanut is a cost-effective nutritional powerhouse due to its high energy value, protein content, and abundance of minerals. In Egypt, there is a growing interest in cultivating peanuts in sandy areas, as they thrive in these conditions. Over half of the peanuts grown are consumed domestically without being processed into oil, while the rest are exported due to their high value in the global market. Peanuts are particularly successful in sandy soils that lack essential micronutrients (Ahmad and Rahim, 2007). However, fungal infections have been observed in this soil. *Fusarium* sp, *Rhizoctonia solani*, *Macrophomina phaseolina*, *Sclerotium rolfsii*, *Aspirigillus* spp, *Pythium* spp and *Melodogyne* spp were isolated from peanut cultivated at the Ismailia Agric. Res. Station (Ismail and Abd El-Momen, 2007).

These fungal diseases, (soil borne fungi) might attach peanut during different growth stages causing considerable losses in the yield (Zhang *et al.*, 2021). Yield losses have been estimated up to 80 %, due to the infection of stem and pods of peanut in the soil and degrades the quality of peanut kernels (Ahmed and Ameen, 2023).

However, these reductions in fungal infection outcomes are no longer known to be effective in preventing fungi, so, biological control presents an intriguing alternative to fungicides for the sustainable management of soil borne diseases (Jacob *et al.*, 2018). Meanwhile, currently, the use of fungicides has been effective in reducing stem rot in peanuts, as many peanut varieties grown are either susceptible or have low levels of resistance. However, the continuous application of fungicides could potentially lead to the development of resistant strains and pose risks to the environment (Bale *et al.*, 2008).

Actinobacteria are a great alternative for controlling plant diseases as they produce compounds that can combat various plant pathogens (Xiao *et al.*, 2002 and Meschke *et al.*, 2012). These bacteria are a diverse

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group found in soil, playing a role in maintaining soil structure, integrity, and nutrient recycling, ultimately benefiting the soil (Ames *et al.*, 1984; Halder *et al.*, 1991 and Elliot & Lynch, 1995). Actinobacteria are widely studied for the production of many economically important antimicrobial metabolites (Lazzarini *et al.*, 2000; Bentley *et al.*, 2002; Saugar *et al.*, 2002; Basilio *et al.*, 2003 and Terkina *et al.*, 2006). Among the various actinobacterial genera, *Streptomyces* spp stand out as they comprise numerous saprophytes, with some even transitioning into advantageous plant endosymbionts. *Streptomyces*' filamentous and sporulating characteristics enable them to thrive in challenging environmental circumstances (Rafik *et al.*, 2014). Certain strains of *Streptomyces* spp have been frequently identified and examined for their potential as biological control agents. These strains are known to be significant producers of bioactive compounds (Berdy, 2012).

Damp environments, crowded plantings, warm weather, and frequent irrigations are believed to be ideal for the spread of diseases in plants. Common signs of disease initiation include the browning and withering of leaves and branches (Koike, 2004). This subsequently affects the yield and its components in peanuts, which makes diseases extremely important. Therefore, the aim of the present work was to replace the chemical disinfectant, along with its potential for bio-control, and its byproducts for controlling soil-borne fungi that cause disease, as well as evaluating seed yield and its various components in the peanut crop under field conditions.

MATERIAL AND METHODS

Genetic Materials:

Two genotypes of peanut (Giza 6 & Introduced 293) were used as genetic materials in the study. The first one is local variety (Giza 6) and the second is introduced from India (Intro. 293). These genetic materials were selected according their economic importance as well as their high susceptibility to soil borne fungi as described by Ismail and Abd El-Momen (2007), both genotypes were obtained from Ismailia Research Station; Oil Crops Section Agricultural Research Center (A.R.C). Name, source, pedigree and growth habit of the studied peanut genotypes are listed in Table (1).

Experimental Field:

A field experiment was carried out in Ismailia Agricultural Research Station, Ismailia Governorate during the two successive summer seasons of 2019 and 2020 using the two peanut genotypes (Giza 6 & Intro. 293).

Table 1. Name, pedigree, source and growth habit of the studied peanut genotypes

Genotype	Pedigree	Source	Growth habit
Giza6	Giza4 x Giza5	Egypt	Erect
Intro. 293	Not available	India	Erect

Soil properties of the Experimental Field:

Biological, chemical properties of the sandy soil were analyzed before sowing. Total counts bacteria, fungi and actinomycetes in the soil samples before cultivation were determined as described by the methods of Clark (1965). Chemical analysis was also, done according to the method outlined by Jackson (1973), as illustrated in Table (2).

Table 2. Chemical and biological properties of experimental soil as an average of the two seasons

Chemical analysis	Total microbial count CFU		
pH	7.98	Bacteria	34x10 ⁴
EC (dS m ⁻¹)	0.93	Fungi	25x10 ³
N (AV %)	11.9	Actinomycetes	23x10 ³
P (AV %)	2.33		
K (AV %)	6.65		

Treatments of the Experimental Field:

Two cultivars were used Giza 6 and Intro. 293 with five treatments, fungicide (Rizolex T50% WP recommended dose) and *Streptomyces erythrogressius* sp (100% -75% - 50% cell suspension filtrate) as well as control treatment (zero) were used in the experiment.

Microorganisms of the Experiment:

The strain used in this experiment *Streptomyces erythrogressius* sub sp. 2, which was isolated from Egyptian soil, identified morphologically in the laboratory and 16sRNA identification at (Sigma Lab, Cairo, Egypt)., submitted in Gene bank with, accession no. LC052669 (Hammad, 2015). The strain was cultured in starch nitrate liquid medium (Tadashi, 1975). The flasks were incubated at 28°C for 7-14 days on incubator checker.

In vitro screening for the active strain for the antimicrobial characteristics against various pathogenic indicator strains, including Gram-negative and Gram-positive bacteria, as well as pathogenic fungi. It shows antimicrobial activity and the active constituent was purified and identified using chemical methods and so assigned But-2-enedioic acid bis-butylamide (antibiotic).

Disinfectant:

Rizolex T 50% WP (Tolclofos methyl) was used as recommended dose (3gm/kg seeds). A fungicides for control of soil borne fungal disease. Seeds were rinsed

multiple times with sterilized water, then immersed in the solution containing bio-control treatments for 30 minutes before being dried in the shade. The seeds were then planted, ensuring that the same concentration of the solution was used for soaking. Each concentration of the organisms was tested in three separate replicates. Control seeds were treated with sterile water. The inocula were cultivated in a liquid medium and combined with living cells and spores suspension before soaking the seeds (Doolotkeldieva *et al.*, 2015).

Experimental design:

Split plot arrangement in a randomized complete block design with three replications in split plot design with the treatments in the main plot and genotypes of peanut in the sub plot was used in this work. Subplot area consisted of three rows each was 2.5 m length, 0.6m width and distance between hills were 20 cm and two seed on hill was carried out.

Agricultural practices management:

The recommended doses of chemical fertilizer (NPK) were applied at sowing by band on one side of the row at 5 cm depth. The recommended agronomic practices such as thinning, manual weeding, irrigation ... etc for peanut plant in plots were done at the optimum time when it needs.

Data Collection and assessment:

- **SPAD chlorophyll content:** Leaf senescence at the flowering stage was assessed by using a hand-held leaf Chl. meter (SPAD-502; Spectrum Technologies, Plainfield, IL) to measure Chl. content on three subsamples from each plant. The Chl. meter provides a measure of the total leaf Chl. content.
- **CO₂ content in soil:** It was determined according to Gaur *et al.* (1971).
- **Total phenol content:** It was estimated in peanut leaves as the procedure given by Zieslin and Ben-Zaken (1993).
- **Seed healthy test:** The experiment followed the guidelines of ISTA. Seeds were placed in pre-sterilized petri dishes with 3 layers of damp blotting paper, with 10 seeds in each dish. All petri dishes were incubated at 25°C for 7 days. Seeds were examined under stereomicroscope and microscope.
- **Peanut characteristic and seed yield:** At harvest, ten guarded peanut plants were randomly chosen from each plot to measure: stem height (cm), number of branches/ plant, pods weight/plant (g), 100- pod weight (g), number of seeds/plant, seed weight/plant (g), 100 seed weight and seed oil percentage (%). According to A.O.A.C. (1990), a sample of 5.0 g from each treatment was used to estimate seed oil %.

Statistical Analysis:

All collected data underwent analysis using analysis of variance (ANOVA) procedure using SAS program and the differences between means using the least significant difference method (LSD) at 5% level of probability was used.

RESULTS AND DISCUSSION

1-Effect of peanut cultivars:

Growth characters of peanut genotypes:

Results in Table (3) indicated that the Intro. 293 was surpassed cultivar Giza 6 in the two seasons, which possessed (48.84 and 50.25) for SPAD values and (56.66 and 64.86) cm for stem height, respectively.

Seed yield and its components of peanut genotypes:

The results in that table showed that Giza 6 cultivar was surpassed by Intro. 293 (84.67) in the number of pods/pl¹ in the second season. Also, it was showed that Intro. 293 was surpassed Giza 6 cultivar in pods weight (154.01 and 185.74)g, 100-pod weight (209.81 and 247.31)g, number of seeds plant (126.42 and 145.33), seeds weight (108.22 and 118.91)g, 100 seed weight (84.15 and 105.3)g and pods yield per faddan (15.73) ardb in the second season, and lower number of pods/pl (80.87) in the second season than Giza 6. The data revealed that there were significant differences between of peanut genotypes under study for some growth characters in Table (3).

In regards to oil percent character, the genotype (Intro. 293) was surpassed cultivar Giza 6 in the two seasons (48.16 and 50.76), respectively. These finding results are agreement with the results detected by Li *et al.* (2019), while it was disagreed with the results reported by Samaha *et al.* (2019) who found that that Giza 6 cultivar surpassed all the tested peanut cultivars in vegetative growth, yield, and its most components traits. In addition, numerous researchers have observed notable differences in the growth, yield, and quality of different types of peanuts, which can be attributed to the varying genetics of the cultivars and how they interact with their surroundings (El-Saady *et al.*, 2014 and Samaha *et al.*, 2019).

2- Effect of bio-agent treatments produced by *S. erythrogriseus* sp 2 on peanut under field conditions in the two seasons.

Growth characters of peanut genotypes:

Results in Table (4) presented the implementation of different concentrations of *S. erythrogriseus* sp 2 varying and adding fungicide Rizolex T 50%WP on SPAD values. These results showed that conc. 50% gave the highest findings, in the first season (48.49) and in the second season (49.72).

Table 3. Impact of two peanut genotypes during the two seasons (S1 and S2) on growth characters, seed yield and its components

Chrac. Treat.	SPAD. Values		Stem height (cm).		Number of pods pl ⁻¹		Pods weight pl ⁻¹ (g).		100- pod weight (g)	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Seasons										
Giza6	41.09	45.28	49.49	52.36	66.54	84.67	114.36	163.16	187.42	240.71
Intro. 293	48.84	50.25	56.66	64.86	77.40	80.87	154.01	185.74	209.81	247.31
LSD.0.05%	0.87	2.52	1.65	2.94	3.14	2.08	3.62	5.44	4.32	6.41

Chrac. Treat.	Number of Seeds pl ⁻¹		Seeds weight pl ⁻¹ (g).		100 seed weight		pods yield ardb fad ⁻¹		Oil -seed percent	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Seasons										
Giza6	93.56	111.60	80.70	96.46	76.73	87.29	11.66	12.75	45.33	48.05
Intro. 293	126.42	145.33	108.22	118.91	84.15	105.37	12.85	15.73	48.16	50.76
LSD.0.05%	2.18	6.33	4.44	3.68	2.34	4.18	N.S	1.36	1.10	1.21

Table 4. Impact of bio-agent *Streptomyces erythrogriseus* sub sp 2 on different concentrations 100%, 75% and 50% and fungicide on growth and yield parameters during two seasons

Chrac. Treat.	SPAD. Values		Stem height (cm).		Number of pods pl ⁻¹		Pods weight pl ⁻¹ (g).		100- pod weight (g)	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Seasons										
Control	41.61	45.55	39.46	48.84	58.0	66.8	104.2	155.8	167.3	221.4
Fungicide	46.48	49.26	53.51	56.99	72.3	77.3	126.0	169.2	196.9	239.8
Conc. 100%	45.06	49.59	62.56	68.01	65.0	72.0	117.1	168.7	199.4	241.1
Conc. 75%	43.18	44.70	55.16	61.33	90.2	110.7	179.6	197.6	214.3	263.0
Conc. 50%	48.49	49.72	54.68	57.87	74.3	87.0	144.2	181.0	215.2	254.8
LSD.0.05%	1.00	1.50	1.19	0.92	1.7	2.3	2.3	1.63	2.28	2.67

The highest result of stem height values 62.56 cm in season 2019 and 68.01cm in season 2020 for stem height treat at conc. 100%. This finding might be because of execution of *B. cereus* 54-1 and *S. erythrogriseus* sp 2 as seed treatment and soil significantly increased chlorophyll content. These findings are in harmony with those reported by Hammad *et al.* (2022).

In addition, in this respect, Manullang and Chuang (2020), studied the impact of *Streptomyces* sp. on *Arabidopsis thaliana* and discovered that the production of IAA by *Streptomyces* sp. was moderate, along with its ability to solubilize phosphate. Inoculation of *Streptomyces* sp. led to an increase in the number of lateral roots, fresh weight, and chlorophyll content in *Arabidopsis thaliana*.

Seed yield and its components of peanut genotypes:

In respect of seed yield and its components of peanut genotypes under different concentrations of *S. erythrogriseus* sp 2 that presented in Table (5) obtained results that the conc.75% gave high values in number of pods (90.2, 110.7), pods weight/pl (179.6 ,197.6)g, number of seeds plant (134.1, 163.2), seed weight/plant (115.2,144.8), 100 seed weight (89.3, 110.6) and pod

yield/feddan (14.62, 17.59) in the first and second seasons, respectively. On the other hand, 100-pods weight, treated with conc. 50% gave high result in first season with value (215.2) g and conc. 75% in the second season with value (263.0) g. According to the above findings, it can say that it's crucial to emphasize that using biological to pre-treat seeds is a viable alternative for controlling severe diseases that can decrease yields by more than 30%, rendering peanut production unprofitable (Paredes *et al.*, 2017). Similar results were obtained by Illa *et al.* (2020) who shows that economically, there are advantages to using *B. subtilis* and *T. harzianum* on peanuts in terms of both improving plant health and increasing productivity. Additionally, it is important to mention that a number of scientists have conducted studies on this topic. Siva *et al.* (2014); Jacob *et al.* (2018) and Samaha *et al.* (2019) showed that these microorganisms are safe and that there are no restrictions in the food codes when their presence is detected on grains.

Table 5. Impact of bio-agent *Streptomyces erythrogriseus* sub sp 2 on different concentrations 100%, 75% and 50% and fungicide on growth and yield parameters during two seasons

Seasons	Chrac.		Number of Seeds pl ⁻¹		Seeds weight pl ⁻¹ (g).		100 seed weight		pod yield ardb fad ⁻¹		Oil -seed percent	
	Treat.	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	
Control		84.0	103.2	64.6	74.6	65.5	76.4	9.70	11.17	45.10	48.36	
Fungicide		98.7	111.8	81.2	96.2	74.5	94.7	11.70	12.81	45.44	47.90	
Conc. 100%		111.0	126.5	113.9	119.9	88.1	96.6	11.29	15.72	47.87	49.86	
Conc. 75%		134.1	163.2	115.2	144.8	89.3	110.6	14.62	17.59	46.84	49.48	
Conc. 50%		122.2	137.7	97.5	102.9	84.8	103.4	13.98	13.91	48.48	51.44	
LSD.0.05%		2.6	5.7	2.1	1.6	1.3	1.7	0.74	0.85	1.00	0.71	

Also, with the same trend, the results showed that the conc. 50% gave high oil percent in the two successive seasons (48.48 and 51.44%) through different concentrations of *S. erythrogriseus* sp 2 (Table 5). Gulluoglu *et al.* (2016) discovered that the oil content of different peanut varieties ranged from 46.96% to 51.55% based on seed dry weight. The cultivar with the highest oil content (51.55%) was Georgia Green, while the lowest (46.96%) was Flower-22.

3-The Interaction effect of bio-agent *S. erythrogriseus* sp 2 in different concentrations on the peanut genotypes in the two seasons:

Growth characters of peanut genotypes:

Results in Table (6) showed that Intro. 293 with conc. 50% gave the heist values in the two seasons (51.6, 55.1), while Giza 6 gave the heist values with the

fungicide on SPAD values in the first season (47.8) and second season (51.3). In the trait stem height, the conc. 100% gave the best result for the genotype except in season 1 (2019) and Giza 6 with the conc. 50% (58.3). These finding results were reported before by Illa *et al.* (2020) who were mentioned that Inoculation of peanut plants with *T. harzianum* and *B. subtilis* in controlled environments demonstrated antifungal effects against the pathogens *A. flavus*, *Fusarium* sp., *S. minor*, and *T. frezzi* at 60 days after sowing. Furthermore, researchers found that the most effective control was achieved when both biological agents were used together. Field trials also showed promising results when applying these bioactive compounds individually or in combination.

Table 6. Interaction between treatments of bio-agent and two peanut genotypes during two seasons and its effect on different concentrations 100%, 75% and 50% and fungicide on growth and yield parameters during two seasons

Seasons	Chrac		SPAD. Values		Stem height (cm).				Number of pods pl ⁻¹			
	Treat.	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	
Control	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293
Fungicide	33.4	49.9	43.4	47.7	38.5	40.4	43.5	54.2	51.7	64.3	74.0	59.7
Conc.100%	47.8	45.2	51.3	47.2	45.6	61.4	48.6	65.4	65.3	79.3	80.3	74.3
Conc. 75%	39.4	50.7	46.4	52.8	57.8	67.4	60.3	75.8	60.0	70.0	78.0	66.0
Conc. 50%	39.6	46.8	41.0	48.4	47.3	63.1	53.0	69.7	87.1	93.3	103.3	118.0
LSD.0.05%	45.4	51.6	44.3	55.1	58.3	51.0	56.5	59.3	68.7	80.0	87.7	86.3
	1.7		3.2		2.3		3.1		4.0		4.0	

Seasons	Chrac		Pods weight pl ⁻¹ (g).		100- pod weight (g)				Number of seeds pl ⁻¹			
	Treat.	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	
Control	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293	Giza6	Intro.293
Fungicide	92.0	116.4	144.6	166.9	153.1	181.4	213.7	229.1	61.7	106.3	91.3	115.0
Conc.100%	108.0	143.9	163.3	175.1	175.0	218.9	242.7	236.8	84.0	113.3	103.0	120.7
Conc. 75%	96.1	138.1	154.7	182.7	186.1	212.8	231.2	251.1	95.33	126.8	108.7	144.3
Conc. 50%	156.0	203.1	182.2	213.0	197.7	230.8	253.4	272.6	120.5	147.7	129.7	196.7
LSD.0.05%	119.7	168.7	171.1	191.0	225.3	205.2	262.7	246.9	106.3	138.0	125.3	150.0
	4.7		5.7		5.2		7.2		4.5		10.4	

Seed yield and its components of peanut genotypes:

Number of pods/plant, pods weight (gm) and number of seeds/plant traits with conc.75% in two genotypes (Intro. 293 and Giza 6) possessed high values in two seasons (2019 and 2020). In trait 100-pod weight Intro.293 and conc. 75% give the highest values results in the 2019 season (230.8) and 2020 seasons (272.6). While Giza 6 possessed give the highest values with the conc. 50% in the both seasons by values 225.3 in first season and in 262.7 the second season (Table 6).

Concerning for seed weight/plant and pod yield/feddan traits, the conc. 75% give the best result for the peanut genotype Giza 6 except in first season with the conc. 100% in the seed weight/plant (117.6)g, while in conc.50% in pods yield/feddan gives 13.5 ardb. For 100 seed weight trait the conc. 75 % give the best values for the genotype Intro. 293 in the second season (118.3 g), while it gave the heaviest seeds (98.5 g/100 seeds) under 100% conc. Increase in root length and shoot length of peanut seedlings may be attributed to the production of such growth regulators, which leads to increase in seed yield and its components. The findings results are in agreements with each of Dochhil *et al.* (2013) and Tančić *et al.* (2013), who are stated that the superior effect of the bio-agents as general trend which positively reflected in growth parameters. The findings in the research conducted by Jacob *et al.* (2016) show that the seedling strength root and shoot lengths were improved by the helpful bacteria *Streptomyces* sp. RP1A-12, which was found to produce indole-3-acetic acid (IAA).

Results in Table (7) showed that Intro. 293 with conc. 75% was possessed the highest values (50.4, 52.4), in the two seasons of oil seed %. While Giza 6 was possessed the highest values (48.3, 50.5) with the conc. 50% in the two seasons (2019 and 2020) in respectively. In this connection, Ismail and Abd El-Momen (2007) reported similarly results during their work in Ismailia with Gregory cultivar gave higher results than Giza 6, they also reported that gypsum addition gives high results.

4- Effect of bio-agent on soil borne fungi infecting peanut under field conditions in two seasons:

The efficiency of different concentrations of bio-agent *Streptomyces erythrogiensis* sub sp 2 in the management of soil borne fungi was assessed on genotypes peanut under field conditions in the two successive seasons. All of the concentrations were suppressed soil borne fungi in different levels of the concentrations compared to control treatment in Fig. (1). Three essential criteria were used to compare the efficacy of testing treatments in season 2021. Based on these criteria, the fungicide treatment Rozelex T 50% WP was the most effective treatment in suppressing most soil borne fungi. In this respect, Jacop *et al.* (2018) reported that the use of crude metabolite formulations led to a significant reduction in disease occurrence. Actinomycetes are known to produce a range of biologically active substances that combat microbial pests.

Table 7. Interaction between treatments of bio-agent and two peanut genotypes during two seasons and its effect on yield and oil percentage parameters

Treat.	Chrac		Seeds weight pl ⁻¹ (g).				100 seed weight			
	Seasons		S1		S2		S1		S1	
	Giza6	Intro.293	Giza6	Intro.293	Giza 6	Intro.293	Giza 6	Intro.293	Giza 6	Intro.293
Control	56.4	72.8	67.3	81.8	58.4	72.6	65.3	87.4		
Fungicide	75.9	86.5	87.4	105.0	66.3	82.7	91.2	98.1		
Conc. 100%	117.6	110.1	106.4	133.5	77.8	98.5	83.8	109.3		
Conc. 75%	90.4	140.2	126.9	162.8	93.5	85.1	102.8	118.3		
Conc. 50%	63.3	131.7	94.3	111.6	87.7	81.9	93.2	113.6		
LSD.0.05%	5.2		4.2		2.9		4.7			

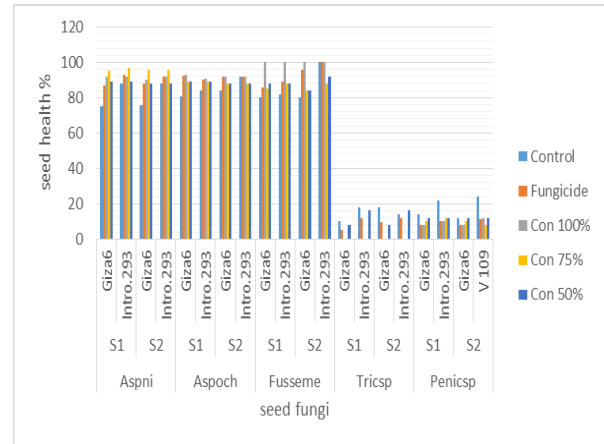
Treat	Chrac.		Pods yield ardb fad ⁻¹				Oil -seed percent			
	Seasons		S1		S2		S1		S2	
	Giza6	Intro.293	Giza6	Intro.293	Giza 6	Intro.293	Giza 6	Intro.293	Giza 6	Intro.293
Control	9.4	10.0	10.2	12.1	44.3	45.9	47.3	49.4		
Fungicide	12.1	11.3	11.4	14.2	44.3	46.6	47.5	48.3		
Conc. 100%	10.7	11.9	13.9	17.6	46.4	49.3	48.4	51.4		
Conc. 75%	12.7	16.6	15.5	19.7	43.3	50.3	46.5	52.4		
Conc. 50%	13.5	14.5	12.8	15.1	48.3	48.7	50.5	52.4		
LSD.0.05%	2.6		NS		1.8		1.5			

These substances, found in crude fractions, are believed to be the key in fighting off pathogens. While previous studies have identified specific metabolites from actinobacteria for controlling plant diseases, the overall effectiveness of utilizing crude metabolite formulations is well-established (Illa *et al.*, 2020).

5- Seeds health test:

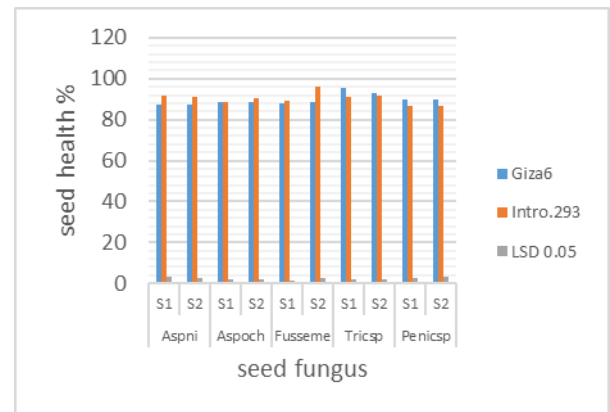
Peanut seeds of all treatments were subjected to health test using blotter method. Application of bio-agent *Streptomyces erythrogienseus* sub sp 2 decreased the severity index to the same level as the control treatment that was not inoculated, as demonstrated in Fig. (1). Overall, in the combined data, lower incidence was recorded in the crude metabolite treatment concentration 100%, which was significantly lower than the un-inoculated control (25%) and disinfectant control (27%). Disease incidence with the two concentrations of bio-agent *Streptomyces erythrogienseus* sub sp 2 filtrate 75% and 50% treatment was also significantly less in compared to the uninoculated control. The peanut Intro. 293 shows less severity of seed born fungi that of cultivar Giza 6 on *Aspergillus niger*, *Fusarium semitectum* in two seasons. While Giza 6 was surpassed in *Trichoderma* sp and *Penicillium* sp in through the two seasons. Also, in concern to *Aspergillus ochraceus*, Giza 6 cultivar was surpassed in season 2019 and Intro.293 in season 2020 as illustrated in Fig. (2). Initial assessments in which various amounts of *Streptomyces erythrogienseus* sub sp 2 filtrate were used to inoculate different peanut cultivars indicated successful suppression of seed-related fungi. The findings illustrated in Fig. (2) underscore that the growth of the *A. flavus* fungus was inhibited., *Alternaria alternaria* and *Cladosporium* sp. were inhibited 100% when *Streptomyces erythrogienseus* sub sp 2 was applied in all concentrations in two seasons (2019 and 2020) compared to control. In this respect, similar results were obtained by Shifa *et al.* (2016), those who have discovered that while fungicides can effectively manage the disease, they also end up raising production costs. However, there is potential for fungicide application to trigger a plant's defense response by using specific chemicals externally, offering protection against *A. alternata* in groundnut crops. This induced response, known as systemic acquired resistance (SAR), has the potential to offer adequate safeguarding against the pathogen, who reported in dual crop trials, they significantly hindered the growth of *A. flavus* by 93-100% and lowered infection rates in both greenhouse and field trials. The authors emphasize the significance of utilizing *Bacillus* to decrease the occurrence of this fungus capable of producing aflatoxins, thus promoting safe peanut cultivation. The inclusion of biological treatments boosted the percentage of plant emergence compared to fungicide treatment, with no substantial

variations observed among the different biological products tested in this study. The percentage was less in case of *Fusarium semitectum*, *Aspergillus niger*, *Aspergillus ochraceus*, *Trichoderma* sp., and *Penicillium* sp, whereas, in case of *Trichoderma* sp by conc. 100% and conc. 75% were inhibited 100%. In the same connection, *Fusarium semitectum*, conc. 100% was inhibited



Aspni: *Aspergillus niger*, Aspoch: *Aspergillus ochraceus*, Fusseme: *Fusarium semitectum*, Tricsp: *Trichoderma* sp., and Penicisp: *Penicillium* sp.

Fig. 1. Impact of bio-agent *Streptomyces erythrogienseus* sub sp 2 on different concentrations and fungicide on incidence and frequency of seed borne fungi of peanut seeds during two seasons compared to control



Aspni: *Aspergillus niger*, Aspoch: *Aspergillus ochraceus*, Fusseme: *Fusarium semitectum*, Tricsp: *Trichoderma* sp., and Penicisp: *Penicillium* sp.

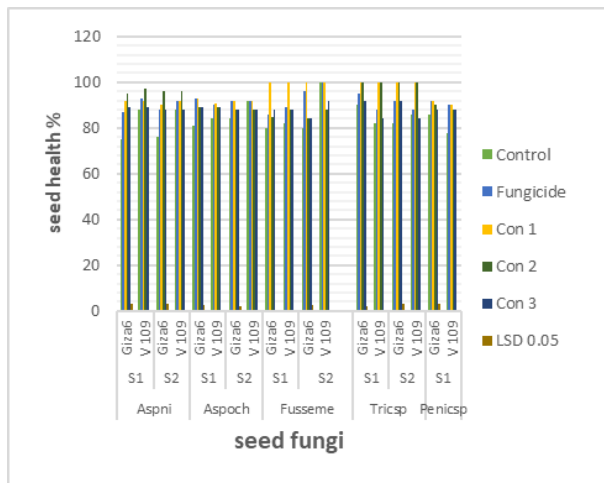
Fig. 2. Impact of two peanut genotypes on incidence and frequency of seed borne fungi of peanut seeds during two seasons

(100%) in two seasons. In *Aspergillus niger*, with conc. 75% was inhibited 96% in two seasons. In *Aspergillus ochraceus*, conc. 100% was inhibited 92%

in the two seasons. In addition, with *Penicilium* sp, conc. 100%, it was inhibited 91% in first season and conc. 75% were inhibited 91% in second season. These findings are agreement with the results detected by Ismail and Abd El-Momen (2007) and Mahmoud (2014) who are use *P. fluorescens* (Pf 5.) in controlling soil borne fungi. While it was disagreement with the results reported by Ahmed and Amein (2023) who use peanut seeds and exposed to microwave and X-ray radiation for controlling the soil borne fungi.

6- The Interaction effect of bio-agent *S. erythrogriseus* sp 2 in different concentrations on the peanut genotypes and its effect on soil borne fungi:

Results in Fig. (3) shows that the cultivars Giza 6 and Intro. 293 with conc. 100% give inhibited the growth 100% of *Trichoderma* sp and *Fusarium semilictum* in the two seasons, followed by *Aspirigillus niger*, which inhibited 97%, in Intro. 293 and con 75% in season 2020, followed by *Aspirigillus ochraceus*, which inhibited 93% in the cultivar Giza 6 with conc.100%, and finally, *Penicilium* sp inhibited 92% in the two peanut genotypes with conc. 100% in through the two seasons (2019 and 2020).



Aspni: *Aspirigillus niger*, Aspoch:*Aspirigillus ochraceus*, Fusseme; *Fusarium semilictum*, Tricsp: *Trichoderma* sp., and Penicisp: *Penicilium* sp.

Fig. 3. Interaction between treatments of bio-agent and two peanut cultivars during two seasons and its effect on frequency of seed borne fungi of peanut seeds during two seasons

Determination of CO₂% and total phenol content:

In the present investigation, accumulation of phenolic content was observed in treated peanut plants. Ground application of *Streptomyces erythrogriseus* sub sp 2 on different concentrations were increased the accumulation of phenolic content in peanut soil rhizosphere. Results in Fig. (4) showed that the levels of

phenolic compounds were further increased, when plants were treated with the bio agents throughout the duration of both seasons. In comparison to the control, *Streptomyces erythrogriseus* sub sp 2 on different concentrations was enhanced the level of phenolic compounds if compared to control.

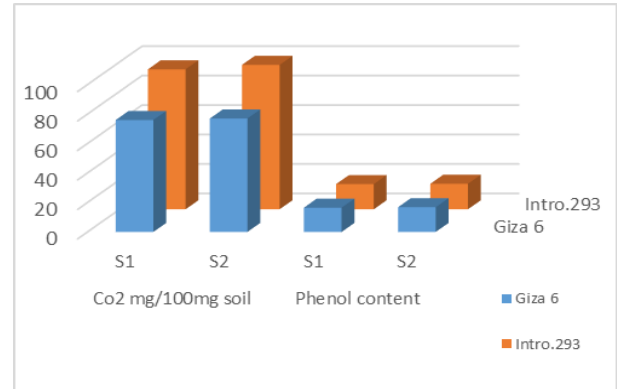


Fig. 4. Impact of two peanut genotypes during two seasons on Phenolic content and CO₂%

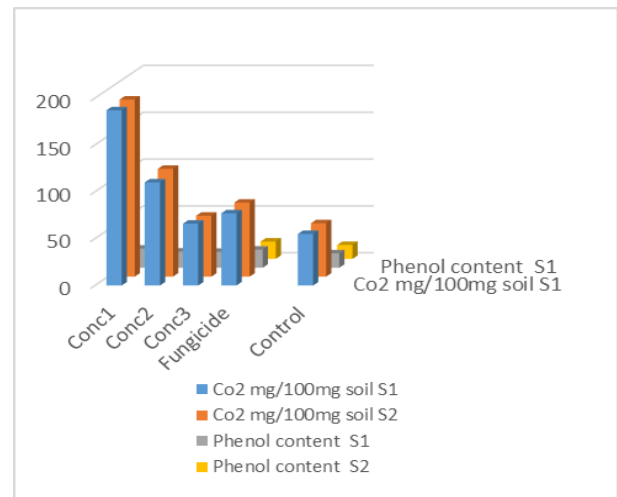


Fig. 5. Impact of bio-agent *Streptomyces erythrogriseus* sub sp 2 on different concentrations 100%, 75%, 50%, fungicide during two seasons on phenolic content and CO₂%

The results show that concentration 100% filtrate give the highest value 20.38 µg/g of fresh weight and the least amount was the control one 14.9 µg/g of fresh weight. The results also were revealed that each of gen.p.293 and Giza 6 give height results in two seasons (Fig. 5). In general, the results showed that the levels of phenolic compounds were further increased when peanut plants were treated with bio-agents throughout both seasons. In comparison to the control, *Streptomyces erythrogriseus* sup sp 2 enhanced the level of phenolic compounds. Phenolic compounds induced lignification in the epidermal regions of plants

as a constitutive and inducible post-penetration method to give resistance against infection (Galeng-Lawilao *et al.*, 2019). Additionally, it was attributed to the activation of the shikimate biosynthetic pathway, which led to the overproduction of metabolites involved in the resistance mechanisms (da Silva *et al.*, 2019).

7-Economic evaluation

Total income, according to MAEBS (2019): Data presented in Table (8) indicated that, the highest value of total income was 16175 and 20034 LE/fed by (Conc. 75%) of Intro. 293 in the first and second seasons, respectively. On other hand, the control in the two cultivars gave the lowest values in total income in both seasons, whereas it gave (9135 and 10414) LE in the first and second seasons, respectively. This economic evaluation was made in this research accordance to Abdel-Azeem *et al.* (2023), whereas it was conducted in this research to know the return and economic return from the nature efficacy of bio-agent in suppressing the soil pathogen fungi and in parallel with evaluation of yield and its components in peanut crop.

Data in Table (8) showed that, the total costs of (control) for two cultivars Giza 6 and Intro. 293 were the lowest costs, it gave 6078 and 5508 LE/fed in the 1st and 2nd seasons, respectively. The cost of treatments (Fungicide) variety cultivar Giza 6 and Intro. 293 were the highest costs, it gave 6978 and 6228 LE/fed in both

seasons. The concentrations (Conc. 75%) of Intro. 293 and the (Conc. 75%) Intro. 293 gave the highest values of net return in first seasons whereas it reaches 9441 and 10593 LE in first and second season, respectively, followed by the concentrations (Conc. 50%) Intro. 293 treatment, it gave 7632 LE/fad. These data as result of final production, whereas increasing total return and decreased disease incidence. These results were in the same line with results reported by Samaha *et al.* (2019). The highest values of benefit/cost ratio 1.40 and 2.11 in the first and second season respectively, it were recorded by (Conc. 75%) and (Conc. 100%) in the first season and second season in respectively. On other hand, the minimum values of benefit /cost ratio were obtained under control and (Conc. 100%) of Giza 6 recording 0.50 and 0.51 in 1st season and 0.86 and 0.86 in the 2nd season, respectively. These data as result of final production which increased total return. Final production was increased because of decreasing disease incidence. These results were in the same line with results reported by which leads to increased income stability and reduce the risk of market glut and falling prices. Intercropping peanut helps to enrich biodiversity, better utilization of environmental resources, and improve farm income. These results were in agreement with Devkota and Nangia (2021).

Table 8. Economic evaluation of Efficacy of bio-agent in suppressing soil borne fungi (Net return and Benefit-cost ratio)

Cultivars	Treatments	Total income (pound. Fed)		Total Cost (Pound/fed)		Net return (Pound/fed)		Benefit-cost ratio	
		S1	S2	S1	S2	S1	S2	S1	S2
Giza6	Control	9135	10414	6078	5508	3057	4906	0.50	0.89
	Fungicide	11807	11593	6978	6228	4829	5365	0.69	0.86
	Conc. 100%	10461	14095	6903	6258	3558	7837	0.51	1.25
	Conc. 75%	12333	15743	6734	6070	5599	9673	0.83	1.59
	Conc. 50%	13123	12987	6515	5883	6608	7104	1.01	1.2
Intro. 293	Control	9769	12305	6078	5508	3691	8614	0.60	1.56
	Fungicide	11017	14461	6978	6228	4039	10422	0.57	1.67
	Conc. 100%	11553	17878	6903	6258	4650	13228	0.67	2.11
	Conc. 75%	16175	20034	6734	6070	9441	10593	1.40	1.74
	Conc. 50%	14147	15316	6515	5883	7632	7684	1.17	1.30

CONCLUSION

Streptomyces erythrograsieus sup sp 2 is effective in controlling peanut soil borne fungi, whereas acting as growth promoter in the current circumstances. Most importantly for a biological control agent used as antifungal metabolites. The findings were confirmed the ability of some tested bio-agents to be near the fungicides efficiency in reducing damping-off, for soil borne fungi diseases as well as increasing the seed yield and its components. So, we recommended to use Intro. 293 peanut variety which superior over Giza 6 in almost all morphological parameters, yield and yield traits.

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الملخص العربي

فعالية بعض العوامل الحيوية فى مقاومة أمراض التربة الفطرية وتأثيرها على البذرة وإنتاجية المحصول فى الفول السودانى

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المستوردة ٢٩٣ وتركيز ٧٥% من العامل الحيوى فى الموسم الثانى. تم اختبار صحة البذرة وثبت أن معاملة النباتات بسلالة *Streptomyces sp* كمقاوم حيوى فى جميع تركيباته قاومت الفطريات الممرضة *Alternaria*، *A. flavus fungus*، *Cladosporium sp.* بنسبة ١٠٠% مقارنة

بالنباتات الغير معاملة وبدرجة أقل لكل من الفطريات

Fusarium semilectum، *Aspirigillus niger*، *Aspirigillus ochraceus*، *Tricoderma sp.*، and *Penicilium sp.* تم عمل اختبار مركبات الفينول وأظهرت النتائج أن

تركيز ١٠٠% من العامل الحيوى أعطى أعلى نتيجة ٢٠,٣٨ ميكروجرام/جرام من الوزن الخضرى. كما تم عمل اختبار نسبة ثانى أكسيد الكربون فى التربة وأظهرت النتائج أن تركيز ١٠٠% أعطى أعلى نتيجة ١٠٠/١٨٧ مللى جم تربة. وقد أوضح التقييم الاقتصادى للتجربة أن التركيز ٧٥% من العامل الحيوى مع السلالة المستوردة ٢٩٣ أعطى أعلى عائد للفدان (١٣٢٢،١٤٤١ جنيه/الفدان) فى الموسم الأول والثانى، على الترتيب.

تم إجراء الدراسة فى حقل للتجارب بمحطة البحوث الزراعية بمحافظة الإسماعيلية أثناء موسمى زراعيين متتالين ٢٠١٩-٢٠٢٠ باستخدام الفول السودانى صنف جيزة ٦ والسلالة المستوردة ٢٩٣. يهدف دراسة تأثير بعض العوامل الحيوية مثل *Streptomyces erythrogressius sp 2* بتركيزات مختلفة من معلق الخلايا (٥٠%-٧٥%-١٠٠%). واستخدام المبيد الفطري (Rizolex T 50% WP). فى مقاومة أمراض التربة الفطرية والتي تسببها بعض أنواع الفطريات مثل (*Fusarium semilectum*، *Aspirigillus niger*، *Aspirigillus ochraceus*، *Tricoderma sp.*، and *Penicilium sp.*).

وتأثيرها على البذرة ونمو وإنتاجية المحصول فى الفول السودانى. وقد أظهرت النتائج أن السلالة المستوردة ٢٩٣ قد تفوقت على الصنف جيزة ٦ فى أغلب صفات نمو النبات وصفات البذرة وإنتاجية المحصول. وقد أوضحت النتائج أن تركيز ٥٠% من العامل الحيوى مع السلالة المستوردة ٢٩٣ أعطت أعلى تركيز من صبغة الكلوروفيل أ (٥٥,١٠). كما سجلت أعلى نسبة من زيت البذرة (٥٢,٤٣%) من السلالة