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Efficacy of biological therapies against onion basal rot caused by *Fusarium oxysporum* f. sp. *cepae* under field and storage conditions

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Abstract

Basal rot disease of onion is caused by *Fusarium oxysporum* f. sp. *cepae* (Hans.) (FOC) economically significant losses wherever onion is grown. *Fusarium oxysporum* were isolated from diseased onion cultivated in different places of Assiut, Egypt. Efficacy of certain yeasts was evaluated for controlling the basal rot of onion *in vitro*. Among of the tested isolates, *Saccharomyces cerevisiae* gave the greatest inhibition (57.74%) and *Candida tropicalis* (1) significantly exerted the greatest reduction of mycelial growth of *F. oxysporum* f. sp. *cepae* (51.18%). Based on the *in vitro* screening, effective yeasts were evaluated under greenhouse, field and storage conditions. Yeasts were applied by two methods (add the pathogen and antagonistic yeasts to soil before sowing seedling onion, and seedling onion soaking in yeasts for 20 minute). Both methods of inoculation showed substantial impact on disease development and plant growth. Add method caused maximum reduction in plant germination, followed by soaking method as compared to control. Application of fungicide (Captain) as compared brought a remarkable increase in seedling emergence of treated plants inoculated with *F. oxysporum* as compared to the untreated plants. In conclusion, tested yeasts were useful as an alternative to chemical control of the onion basal rot and to enhanced growth and yield of onion.

Keywords: *Allium cepa*, basal rot, antagonism, *Fusarium oxysporum* f. sp. *cepae*, yeasts.

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1. Introduction

Onion (*Allium cepa* L.), is one of the oldest vegetables, has been used as spice and medicine for thousands of years (Keusgen, 2002). *Fusarium* basal rot of onion is an economically important disease to which onion bulbs and shallots are sensitive during all their growth stages (Cramer, 2000). To manage the disease, chemical control is very effective, but it is not economical and pollutes the environment. Use of resistant cultivars is another acceptable strategy of control however onion cultivars with acceptable level of resistance are limited. Researchers have recently considered biological control as a complementary approach for controlling this disease. Yeasts treatments were suggested to play a beneficial role in cell division and cell enlargement (Freimoser et al., 2019). yeasts were demonstrated to be effective biocontrol agents of seedlings onion post-emergence damping off. Various yeast species have been reported as active biological control agents. Some effectively *saccharomyces cerevisiae* was used as bioagents root rot pathogens under greenhouse conditions (Abd El-kader et al., 2012). *Candida glabrata* and *C. maltosa* significantly reduced the incidence of late maize wilt disease when applied by seed inoculation (El-Mehalawy et al., 2004). *Pichia guilliermondii* gave hence possess potential to control wilt disease in tomato crop (Nguyen et al., 2011). Shalaby and El-Nady (2008) found that seed soaking, or soil inoculation with *S. cerevisiae* increased germination rate, survival of plants and reduction of pre and post emergence damping off and inhibited *Fusarium oxysporum* liner growth *in vitro*. Also, they found that pre- and post- emergence damping off was reduced significantly when seeds of faba bean were coated with a water suspension of the yeast (10^9 CFU ml^{-1}). The beneficial effects of these microorganisms last longer than that of chemicals and can therefore protect the plant throughout all growth stages. Under greenhouse and field conditions, control of plant diseases using antagonistic yeasts can be effective. Also, biological control can limit the instances of basal rot of onion caused by *F. oxysporum* and reduced probability of disease development. If pathogen attacks the host plant late in the season, the symptoms may not appear until onion bulbs are in storage (Ozer et al., 2003) Present study found that biological agents inhibit the growth of *F. oxysporum. Fusarium oxysporum* f. sp. *cepae* (Hanz) Snyd attack onion bulbs in storage and cause rotting of onion bulbs during storage. In storage, onion bulbs appear spongy or sunken, infected bulbs are softened, brown and watery when cut open during storage bulbs are affected by many microorganisms leading to rot being commercially important crop of the state; it was felt necessary to carry out investigations on storage rot of onion. Despite the achievement in production technology and availability of good varieties of onion, the post harvest losses during storage is still an ailing cause which leads to significant qualitative and quantitative losses during storage up to 25-30%. The onion postharvest losses were estimated worth Rs 600 crores is found to be due to desiccation, decay and sprouting, (Kumar et al., 2015). The rationale behind such post-harvest losses till today is the unavailability of good storage facilities during post-harvest storage phase. There seems a big gap between the storage facility and the storage capacity which is ultimately leading to the unforeseeable postharvest decay and deterioration of onion bulbs. Therefore, the main objectives of this study were attempted to apply a of some antagonistic yeasts, enhanced the biocontrol of onion basal rot and studied the suppressive effect of some free yeast strains against *Fusarium oxysporum*, under greenhouse, field and storage conditions.

2. Materials and methods

2.1 Isolation of the causal pathogen of onion basal rot disease

Bulb samples of onion showing typical basal

rot symptoms were collected from different locations in the Assiut governorate of Upper Egypt. Isolation of the pathogen from the infected bulbs was performed by the tissue segment method described by Rangaswami (1958). Infected portions of bulb samples were cut into small pieces and washed thoroughly in tap water. The pieces were then disinfected by immersing in 1% sodium hypochlorite (SH) solution for one minute, rinsed three times in sterilized distilled water (SDW), and dried between folds of sterilized filter papers. Disinfected pieces were transferred aseptically into Petri plates containing Potato Dextrose Agar (PDA) medium supplemented with 400 mg streptomycin sulfate per liter of medium. The plates were then incubated at 25±0.5 °C for 5 days and examined daily for fungal growth. The growing fungal colonies were purified by single spore and hyphal tip techniques, followed by sub-culturing onto a freshly prepared PDA medium at the same conditions. According to cultural and microscopical characteristics of colony mycelia and spores, isolated fungi were morphologically identified (Leslie & Summerell, 2006; Gerlach & Nirenberg, 1982). Pure cultures of isolated fungi were maintained at 5 °C on PDA slants for further studies.

2.2 Pathogenicity test

The pathogenic capability of all fungal isolates obtained to cause basal rot disease was investigated on the onion Giza 6 cultivar under greenhouse conditions. The inoculum of each isolate tested was prepared by placing two disks (0.7 cm in diameter) taken from the 7-day-old culture onto an autoclaved sand-maize medium (80 g sieved fine river sand, 20 g maize meal, and 50 ml water) in conical flasks tightly closed with cotton plugs. Then flasks were incubated at 27±0.5 °C for 14 days. Formalin-sterilized plastic pots (35 cm in diameter) were filled with autoclaved loam soil (5.0 kg of each), infested with 150 g inoculum of each isolate tested, and then slightly irrigated every other day for a week. Pots treated with the same amount of non-inoculated sand-maize medium served as control. Another week later, onion seedlings were disinfected by dipping in 1% SH solution for 3 min, rinsed three times in SDW for 10 min, and then transplanted at a rate of 6 seedlings per pot. The experiment was performed with four pots (replicates) of each isolate tested in a completely randomized design. Then pots were checked daily and irrigated when necessary. Observations of basal rot symptoms were recorded daily and continued with disease development until the whole plant's complete rotting. Six months after planting, plants were uprooted, and percentages of disease incidence (DI) and surviving plants (SP) were calculated according to the following formula:

$$DI\ \% = \frac{Number\ of\ diseased\ plants}{Total\ number\ of\ plants} \times 100$$

$$SP\ \% = \frac{Number\ of\ surviving\ plants}{Total\ number\ of\ plants} \times 100$$

2.3 Preparation and growth conditions of used yeast strains

Pure cultures of yeast strains were previously isolated from different locations in Egypt and identified according to classical methods used in yeast taxonomy (van der Walt & Yarrow, 1984). Yeast strains which exhibit the highest antagonistic effects against the pathogenic *F. oxysporum* were selected and cultured on glucose peptone yeast extract (GPY) medium (Difco, 1985) by shaking on a rotary shaker at 28±0.5 °C for two days.

2.4 Study of antifungal activity of yeast strains against FOC

Nineteen yeast strains were tested for the antifungal activity against FOC using the direct dual culture technique. Yeast strains were initially grown on GPY broth in test tubes by shaking on a rotary shaker at 28±0.5 °C for 2 days, as mentioned before. On the GPY plates' surface, 0.1 ml of each aqueous yeast suspension with a concentration of 10^6 cells ml^{-1} were plated. Then yeasts were grown for 5-7 days as a lawn on the Sabouraud agar surface, whereby 5 mm discs in diameter were cut. Disks from each yeast strain were placed on a PDA medium on one side of the Petri plate, and the opposite side at an equal distance was inoculated by 5 mm- disks of FOC isolate taken from 7-days-old culture. Four plates were used for each yeast strain tested, and the inoculated plates with FOC discs only were served as control. The entire experiment was twice repeated for confirmation of the results. After 10 days of plate incubation at 28±0.5 °C, antifungal activity was determined by measuring the zone of inhibition (cm) produced by yeasts against FOC. Also, the located mycelium inside the zones of inhibition was microscopically examined. Growth inhibition (%) of FOC was calculated using the following formulae:

$$GI\ \% = \frac{C - T}{C} \times 100$$

Where GI = growth inhibition, C = mycelium radial growth in control, and T = mycelium radial growth produced by yeast strain tested.

2.5 Evaluation of antagonists under greenhouse conditions

Yeast strains that showed the highest suppression rate against FOC *in vitro* were applied to control the disease under greenhouse and field conditions. Pots of 35 cm diameter were filled with sterilized soil mixed with the FOC inoculum one week before planting. Soil infestation was conducted 7 days before sowing by adding 3% of the inocula with the soil in each pot. All treatments were applied by two methods the first was add the pathogen and antagonistic fungus (yeasts) at the rates of 10^6 CFU/g were mixed with soil pots before sowing seedling onion, the second methods seedling onion soaking in yeasts for 20 minutes. Seedling onion treated in fungicide Captain (50% WP) was used as a reference. Each pot was planted with 5 seedling onion Giza 6 cultivar. Four replicates were used to each treatment and pots containing inoculum of FOC only were used as control. Plants were irrigated when necessary and examined periodically. Data disease incidence and plant mortality were recorded after 6 months of seed sowing. The formulae used to determine the diseases incidence were followed as mentioned before.

2.6 Evaluation of antagonists under field conditions

A field experiment was conducted to evaluate the efficacy of biocontrol agents on onion basal rot during 2019/2020 and 2020/2021 seasons. Seedlings of onion cv. Giza 6 were planted in rows in plots size 5 × 4 m and spacing 15 cm. A Randomized Block Design (RBD) was used with three replications. Before sowing, the treatments were applied by two methods such as greenhouse treated, and fungicide Captain was used for comparison. All normal agronomical practices including irrigation, weeding and fertilizer application were followed at regular intervals. Natural incidence of basal rot of onion was recorded. At harvest time, plant samples (10 healthy plants each) were taken at random from each plot to determine the growth parameters, plant

height (cm), bulb diameter (cm), Average bulb weight (g) and total bulb yield (ton/feddan) were recorded (feddan = 4200 m² = 0.420 hectares = 1.037 acres).

2.7 Bulb storage

After harvest, 5 kg onion bulbs for each replicate from treatments were placed in nylon mesh onion bags without removing the stalks and Store in a well-ventilated place. During storage the air was continuously drawn over the onions by means of a fan to remove excess moisture. The bulbs were stored at low temperature for 30, 60, or 90 days. After the appropriate duration of storage, five bulbs were removed from each replicate bag and evaluated for severity of Enterobacter bulb decay. Each onion bulb was sliced from the neck to the basal plate, and the cut surface area of the fleshy scales was rated visually for severity of Enterobacter bulb decay (percentage of the surface area of the fleshy scales showing symptoms typical of this disease) (Schroederand du Toit, 2010). A low incidence of bulbs (1 to 5%) showed symptoms characteristic of other storage rots (e.g., neck rot caused by *Botrytis aclada* and *B. allii*, black mold caused by *Aspergillus niger*, Fusarium basal rot caused by *Fusarium oxysporum* f. sp. *cepae*.

2.7 Statistical analysis

Analysis of variance (ANOVA) was carried out using MSTAT-C program. The least significant difference (LSD) at P≤0.05 was applied to detect differences among treatments (Gomez and Gomez, 1984).

3. Results and Discussion

3.1 Pathogenicity test

Nine isolates of *Fusarium oxysporum* f. sp. *cepae* were isolated from different localities of Assiut governorate in Egypt. Identification of the isolates was based on spore morphology and culture characteristics, as described by Booth (1985) and Schwartz (2010) (Figure 1).

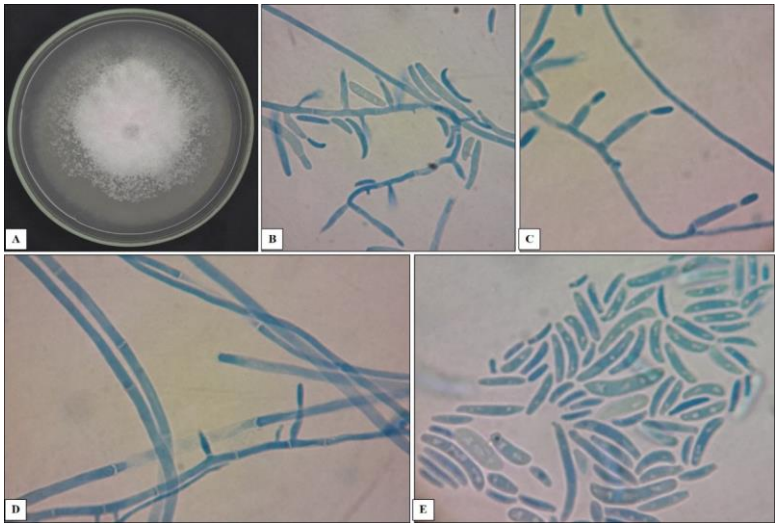

Figure 1: *Fusarium oxysporum* f. sp. *cepae*; A: colony on DPA after 7 days, B, C and D: hyphae bearing short monophialides, E: micro- and macroconidia.

Pathogenicity test of isolates are presented in Table (1) indicate that all isolates proved to be able to infect onion plants causing basal rot symptoms. Typical symptoms observed were

yellowing and eventual dieback of the leaves, drying of leaves and in advance stage, basal portion of plants completely rotted and entire plant got collapsed on ground, which was noticed 15 days after transplanting in the pots. Uninoculated plants remained healthy. These results agree with Saxena (2007) isolated *Fusarium oxysporum* f. sp. *cepae* from infected bulb in USA. Davis (2008) reported the pathogen, *F. oxysporum* f. sp. *cepae* causing onion and garlic basal rot. Dissanayake et al. (2009) studied the pathogenicity of 32 isolates of *Fusarium* spp. on five commercial cultivars of welsh onion and proved that five *Fusarium oxysporum* isolates had higher virulence to *Allium* spp.

Table 1: Pathogenic capability of *F. oxysporum* f. sp. *cepae* on onion cultivar Giza 6.

Isolates number	Disease incidence (%)	Surviving plants (%)
1	41.66	58.33
2	45.83	54.16
3	50.00	50.00
4	66.66	33.34
5	29.16	70.83
6	33.33	66.67
7	37.49	62.50
8	20.82	79.17
9	79.16	22.22
Control	0.00	100.00
L.S.D. at 5%	11.41	2.77

3.2 Evaluation of yeast isolates as biocontrol agents against the causal pathogen *in vitro*

Yeast isolates from different sources were screening for their abilities to inhibit the growth of the plant pathogen *F. oxysporum*. These isolates were *Candida tropicalis*, *Cryptococcus albidus*, *Debaryomyces hansenii*, *D. pseudopolymorphus*, *Galactomyces candidus*, *G. pseudocandidus*, *Klyuveromyces marixianus*, *Lachancea thermotolerans*, *Papiliotrema laurentii*, *Pichia caribaea*, *Saccharomyces cerevisiae*, and *Saccharomyces* sp., which were identified by using morphological and physiological characters according to Barnett et al. (2000) and molecular by the internal transcribed spacer (ITS) sequences of nuclear ribosomal DNA. The studied yeast nineteen isolates were shown variable antagonistic impact against the plant pathogen *F. oxysporum* f. sp. *cepae* giving inhibition percentages of the pathogen growth ranging from 22.28 to 57.74 (Table 2 and Figure 2). The strains *Saccharomyces cerevisiae*, *Candida tropicalis*, *Pichia caribaea* and *Saccharomyces* sp. were exhibited the highest antagonistic effect. These promising strains were selected for the applied experimental studies in greenhouse and field.

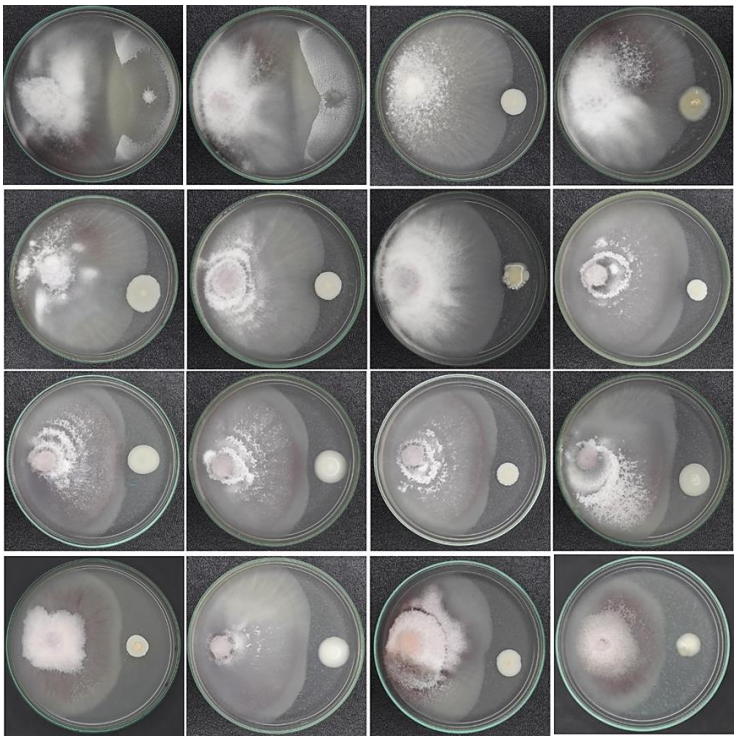

Figure 2: Antagonistic effect of different antagonistic yeasts against *Fusarium oxysporum* f. sp. *cepae* on PDA at 28 °C after 10 days.

Results are presented in Table (3) and Figures (3 and 4) reveals that linear growth of *Fusarium oxysporum* f. sp. *cepae* was significantly decreased at all the tested treatments. The highest decreased in linear growth was pronounced by *Saccharomyces cerevisiae* and *Candida tropicalis* (1) by (57.74% and 51.18 %) respectively.

Table 2: Effect of certain yeasts on inhibition growth of *Fusarium oxysporum* f. sp. *cepae in vitro*.

Yeast	Inhibition growth (%)
Control	00.00
Saccharomyces cerevisiae	57.74
Candida tropicalis (1)	51.18
Candida tropicalis (2)	46.29
Candida tropicalis (3)	42.59
Cryptococcus albidus	34.81
Saccharomyces sp.	33.33
Lachancea thermotolerans	31.85
Pichia caribaea	35.19
Saccharomyces sp.	31.85
Saccharomyces paradaxus	36.96
Saccharomyces cerevisiae	30.37
Papiliotrema laurentii	30.37
Candida tropicalis	28.89
Debaryomyces hansenii	28.89
Debaryomyces pseudopolymorphus	27.41
Klyuveromyces marixianus	27.41
Galactomyces candidus	24.44
Galactomyces candidus	23.70
Galactomyces pseudocandidus	22.22
L.S.D. at 5%	4.21

Table 3: Effect of selected antagonistic yeasts against *Fusarium oxysporum* f. sp. *cepae in vitro*.

Yeast	Inhibition growth (%)
Control	00.00
Candida tropicalis (1)	51.18
Candida tropicalis (2)	46.26
Candida tropicalis (3)	42.59
Saccharomyces paradaxus	36.96
Saccharomyces cerevisiae	57.74
Pichia caribaea	35.19
L.S.D. at 5%	3.43

The *Pichia caribaea* gave the least decreased by 35.19%. The mycelial growth of *Fusarium oxysporum* f. sp. *cepae* was influenced and much reduced by other treatments. This agreed with data obtained by Astasa and Aliad (2005) who reported that 4 yeast isolates showed some inhibitory effect on the growth of *F. oxysporum in vitro*.

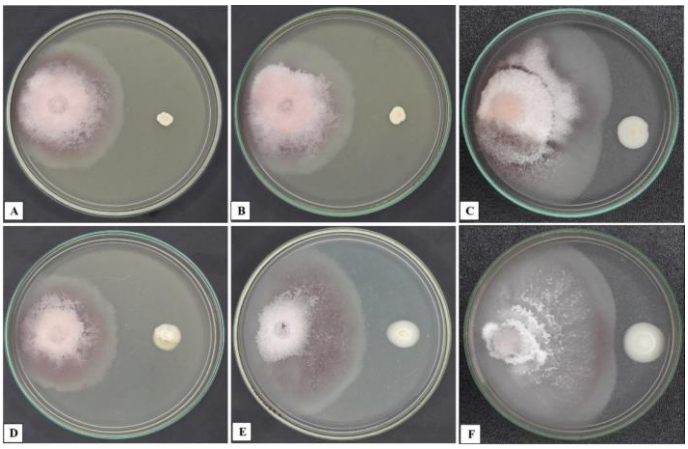

Figure 3: A, B and C: Antagonistic effect of *Saccharomyces cerevisiae* against *Fusarium oxysporum* after 5, 7 and 10 days on PDA medium. D, E and F: Antagonistic effect of *Candida tropicalis* against *Fusarium oxysporum* after 5, 7 and 10 days on PDA medium.

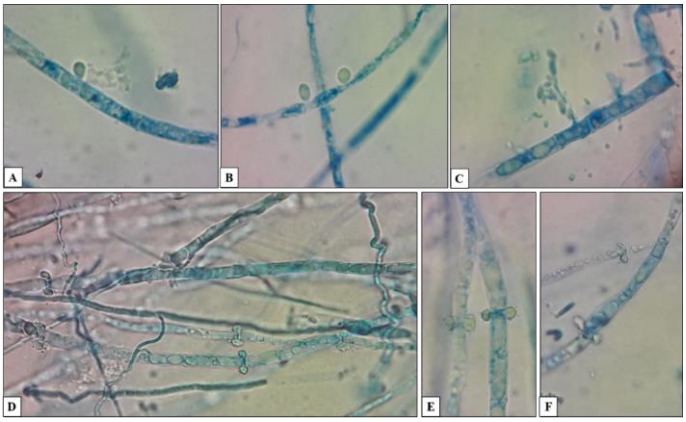

Figure 4: Interaction of yeast with the pathogenic *Fusarium oxysporum* in dual plate confrontation. A, B and C: yeast cells attack the pathogen hyphae. D, E and F: Papillae-like structures.

Yeast isolates which were found to be strongly antagonistic to *Fusarium oxysporum* f. sp. *cepae in vitro* were effective producers of antifungal metabolites (El-Mehalawy et al., 2004). In addition, it was found that the isolates of actinomycetes produced chitinase and β-1, 3- glucanase and caused extensive plasmolysis and cell wall lysis of *Fusarium oxysporum* f. sp. *cepae in vitro*. Also, a yeast's mechanism for biocontrol involves nutrient competition and by-production of extracellular substances in the wound site of the host that causes collapse and degradation of the fungal hyphae (Baker & Cook, 1974).

3.3 Evaluation of antagonists under greenhouse conditions

In pot experiment, all antagonists reduced the disease severity. Table (4) showed that under greenhouse conditions the percentage of basal rot disease incidence in first method recorded between 0-26.66% but second methods in ranged between 6.66-33.33%. Both *Saccharomyces cerevisiae* and *Candida tropicalis* (1) had greater decreased in diseases caused by *Fusarium oxysporum* f. sp. *cepae* by 0.00, 6.66, 6.66 and 6.66% in two methods respectively followed by *Candida tropicalis* (3) and fungicide (Captain) by 13.33, 13.33 and 20.00%. These results agreed with El-Mehalawy *et al.* (2004) found that the production of root and attached soil particles of 14-day-old onion seedlings were colonized to different degrees by yeast isolates with the frequency of colonization being significantly ($P < 0.05$) greater in the first 2 cm of root and soil. Root colonization frequency in the rhizosphere soil was greater in treated plants by yeasts.

Table 4: Effect of antagonistic yeasts on the incidence of onion basal rot diseases under greenhouse conditions.

Yeast	Disease incidence (%)		Surviving plants (%)	
	Add	Soaking	Add	Soaking
Candida tropicalis (1)	6.66	6.66	93.33	93.33
Candida tropicalis (2)	20.00	33.33	80.00	66.66
Candida tropicalis (3)	13.33	13.33	86.66	86.66
Saccharomyces paradaxus	20.00	20.00	80.00	80.00
Saccharomyces cerevisiae	0.00	6.66	100	93.33
Pichia caribae	26.66	40.00	73.33	60.00
Fungicide (Captain)	13.33	20.00	86.66	80.00
Control	80.00	80.00	20.00	20.00
L.S.D. at 5% (Treatments)=	17.35		22.44	
(Methods) =	6.36		5.58	
(Treatments × Methods) =	n.s		n.s	

3.4 Evaluation of antagonists under field conditions

Treatments that contained yeasts significantly ($P<0.05$) reduced the incidence of onion basal rot disease. There were significant differences in the disease index, the number of diseased onion seedlings between two methods (add the pathogen and antagonistic fungus in soil and seedling onion soaking in yeasts). The application of each antagonistic yeast using add method increased the percentage of onion basal rot control. Table (5) shows the effect of antagonistic yeasts and fungicide Captain on onion basal rot disease under field conditions and all used treatments significantly reduced the disease. Data also show that the highest values of decrease occurred with fungicide (Captain) and *Saccharomyces cerevisiae*, followed by the treatments of *Candida*

tropicalis (1) and *Candida tropicalis* (3), while *Pichia caribae* and *Candida tropicalis* (2) recorded the lowest reduction compared with untreated plants (control) in two seasons. Such results are in accordance with that reported by Kamel et al. (2016) found that yeast strains isolated against *Fusarium oxysporum* f. sp. *cucumerinum* causal agent of cucumber wilt disease in soil. Also, antagonists were applied by soil infestation as in a previous work (Zeidan et al., 2018). The main mechanism involved in late basal rot disease reduction by the yeast fungi is the production of non-volatile diffusible metabolites since the production of these metabolites was related to significant in vitro inhibition and biological control of the pathogen. Once cell wall damage has occurred, the pathogen is more likely to be susceptible to attack by other biological, physical and chemical agents. The antagonistic yeasts in the present study were capable of growing totally at the expense of the hyphae of Fusarium, indicating their potential for pathogen suppression (Joubert and Doty, 2018) where the antagonism takes place outside the limits of rhizosphere.

Table 5: Effect of antagonistic yeasts on the incidence of onion basal rot diseases under field conditions.

Yeast	Disease incidence (%)			
	2019/2020		2020/2021	
	Add	Soaking	Add	Soaking
Candida tropicalis (1)	11.33	12.00	11.16	13.66
Candida tropicalis (2)	19.33	21.66	20.00	20.33
Candida tropicalis (3)	12.00	12.88	12.66	13.33
Saccharomyces paradaxus	13.5	15.33	12.66	16.00
Saccharomyces cerevisiae	10	10.16	9.66	10.66
Pichia caribae	14.66	15.66	14.16	16.83
Fungicide (Captain)	10.5	9.5	8.16	9.33
Control	78.33		79.00	
L.S.D. at 5% (Treatments)=	1.64		1.78	
(Methods)=	0.399		0.28	
(Treatments × Methods)=	1.12		1.86	

3.5 Evaluation of antagonists on yield

Data presented in Table (6) indicate that the tested sowing dates affected, plant height (cm), bulb diameter (cm), average bulb weight (g) and total bulb yield (ton/feddan) in the two tested growth seasons. Onion growth measurements were data in first method best than second method compared control in two seasons. Values of parameters of onion plants were significantly increased with the dual inoculation each isolates yeast fungi. The increase of plant growth could be attributed to the role yeasts present in inocula. Soil inoculation with *Saccharomyces cerevisiae* and fungicide gave higher records of all parameters followed by *Candida tropicalis* (1), and *Candida tropicalis* (3). The increase of onion resistance obtained in this study, could be related to the role of yeasts as plant growth promoters. The yeast fungi producing growth. These metabolites are considered important after being taken up by the plant or indirectly by modifying the rhizosphere environment. Höflish and Kühn (1996) reported that the promotion of cruciferous oil and intercrops and nutrient uptake was stimulated by inoculating rhizosphere yeast fungi. Also, rhizosphere yeast fungi promoted plant growth by oxidizing ammonium to nitrate, oxidizing elemental sulphur to sulphate and solubilizing insoluble phosphate.

Yeasts may therefore prevent this growth inhibition, and generally increase the plant's tolerance to stress. These enzymes may also serve as a mechanism for ammonia secretion by the yeasts, which have been reported in the past and could serve as a way for the plant to recycle nitrogen using its symbiotic partners (Dewedar and Ibrahim, 2016).

Table 6: Effect of antagonistic yeasts on growth parameters of onion plants during 2019/2020 and 2020/2021 seasons.

Yeast	Methods	2019/2020				2020/2021			
		Plant height (cm)	Bulb diameter (cm)	Average bulb weight (g)	Total bulb yield (ton/feddan)	Plant height (cm)	Bulb diameter (cm)	Average bulb weight (g)	Total bulb yield (ton/feddan)
Candida tropicalis (1)	Soaking	64.66	3.90	67.50	15.83	64.83	4.10	65.33	15.00
	Add	66.33	4.56	72.00	16.50	66.83	4.56	71.00	16.00
Candida tropicalis (2)	Soaking	61.00	3.63	62.00	13.16	61.16	3.76	63.00	12.83
	Add	62.50	3.96	62.16	14.00	62.83	4.23	62.83	13.5
Candida tropicalis (3)	Soaking	62.16	3.70	64.33	14.50	62.33	3.93	65.5	15.16
	Add	64.00	4.26	66.33	15.00	64.83	4.13	67.16	15.83
Saccharomyces paradaxus	Soaking	60.33	3.60	62.16	13.00	60.00	3.83	62.00	13.50
	Add	61.00	4.30	63.66	13.00	61.50	4.53	65.16	14.83
Saccharomyces cerevisiae	Soaking	66.33	5.56	68.16	16.33	67.33	6.10	68.5	16.00
	Add	68.16	6.23	74.16	16.50	68.66	6.35	75.16	17.00
Pichia caribae	Soaking	50.66	3.50	61.00	10.66	51.00	3.50	61.33	12.00
	Add	55.66	3.90	62.16	11.50	57.00	3.93	63.00	12.83
Fungicide (Captain)	Soaking	57.16	5.20	66.16	15.33	57.00	5.53	65.66	14.33
	Add	58.66	5.16	70.66	15.00	57.83	5.50	69.83	16.00
Control		45.33	3.10	56.33	6.50	48.00	3.36	55.16	5.66
L.S.D. at 5% (Treatments) =		2.51	0.23	1.69	1.46	1.46	0.46	1.01	0.59
(Methods) =		1.11	0.12	1.02	0.41	0.67	0.22	0.63	0.45
(Treatments × Methods)=		3.13	0.34	2.64	1.17	1.91	0.62	1.77	1.27

3.6 Bulb storage

Inoculated bulbs stored for 1 or 2 months after curing did not differ significantly in severity of bulb rot, but bulb rot was significantly more severe for bulbs stored for 3 months after curing postharvest disease incidence was greatly effective. Under storage house conditions, all treatments decreased the incidence of basal rot diseases compared to control as reported in Table (7). The results showed which obtained by visual observation that after storage for 3 months, the treatments were effect on diseases.

Table 7: Effect of treatment by of yeast on fungal disease incidence of onion at storage house.

Yeast	Number of infected onions in storage house			
	2019/2020		2020/2021	
	Add	Soaking	Add	Soaking
Control	40	40	44.33	44.33
Candida tropicalis (1)	2.66	2.66	3.06	3.00
Candida tropicalis (2)	3.06	4.06	3.96	3.16
Candida tropicalis (3)	2.66	2.66	3.4	3.66
Saccharomyces paradaxus	2.13	2.33	3.33	3.16
Saccharomyces cerevisiae	2.66	3.33	2.5	3.00
Pichia caribae	3.5	3.76	3.66	3.66
Fungicide (Captain)	4.00	4.13	3.5	4.63
L.S.D. at 5% (Treatments) =	2.93		5.59	
(Methods) =	0.92		0.65	
(Treatments × Methods) =	2.62		1.85	

These results confirm that *Saccharomyces cerevisiae* treatment can result between in a 97.5 and 97% reduction in fungal damage on onions during storage compared (60 and 54.66%) onions in the untreated control group were infected by basal rot diseases. Storage is one of the important aspects for post-harvest handling of onion (Anbukkasari, 2013). The storage condition extends the period of availability of fresh onion by arresting the metabolic breakdown and decay. It is achieved by controlling the physiological activity, biochemical activity, microbial invasion. Inadequate and improper field curing after harvest, infection by different pathogen. In general, the losses, currently about 35-40% of the onion is estimated to be lost as postharvest losses during various postharvest operations including storage.

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