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# Protective Foliar Approaches against Downy and Powdery Mildews of Cantaloupe under Plastic Houses Conditions

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***Abstract*—Evaluating the efficacy of different plant resistance inducers and/or bio-agents treatments against foliar diseases incidence revealed that plant spray with treatments, [Calcium chloride + S. cerevisiae + Chitosan] and [Potassium bicarbonate +Thyme oil] resulted in the highest reduction in foliar diseases Downy and Powdery mildews diseases incidence and severity on cantaloupe plants comparing with fungicides treatment was carried out under commercial plastic houses conditions. The applied foliar treatments could highly reduce the incidence of both downy and powdery mildews as well as suppress the development of both diseases expression comparing with fungicide treatment. The obtained results in the present study has shown the potential of tested some plant resistance inducers as effective inhibitors against downy and powdery mildews when used alone or combined with antagonistic bioagent S. cerevisiae.***

***Index: Calcium chloride, Cantaloupe, Chitosan, Downy mildew, Thyme oil, Fungicides, Plant Resistance Inducers, Potassium bicarbonate, powdery mildew, S. cerevisiae.***

## I. INTRODUCTION

Cantaloupe (*Cucumis melo* L. var. *reticulates* Ser.) is one of the most popular cucurbit crops in Egypt either for local consumption or exportation. Downy and powdery mildews are most destructive to cucumber and cantaloupe, though all cucurbits are susceptible. Although powdery mildew has a long history in greenhouse production, downy mildew has recently been the focus of attention and concern. Both diseases can contribute to significant economic losses in many greenhouse horticultural crops.

Although the two disease share the name "mildew', they are very different. Powdery mildew infections reduce crop aesthetics and value but usually do not result in plant death. In contrast, downy mildew infections often result in plant death as well as the loss of aesthetics [1]. Downy mildew caused by the fungus like *Pseudoperonospora cubensis* is among the most serious diseases affecting cantaloupe production, with a high severity, especially at the time of fruit maturity [2,3]. Meanwhile**,** powdery mildew caused by *Sphaerotheca fuliginea* is a very common disease that can reduce yield (fruit quantity and/or size) and market quality (flavor, color, storability, etc) in melons [4,5].

It is well known that Successful control of downy and powdery mildews using chemical control of plant diseases greatly cause environmental pollution and increase in the accumulated toxic substances in human food chain. On the other hand, using other trials of disease management, *e.g.* biological control, plant extracts, antioxidants and agricultural practices, could give enough efficient results**.**

The best way to ensure success of a disease- management program is to use integrated disease-control measures [6]. Generally, IPM is regarded as the use of environmentally safe practices to reduce the disease incidence and development or use of multiple control tactics integrated into a single pest control strategy [7]. For example, different natural products, *i.e*., biocontrol agents, plant extracts and natural compounds were used as an IPM program to control powdery mildew of greenhouse crops [6,8]. Since economic thresholds have not been established for most plant pathogens, an IPM takes a somewhat different approach in plant disease control. Salts have been previously studied as foliar applied control agents for powdery mildews on various horticultural crops. A recent review [9] discussed the use of different foliar applications including compost or plant extracts, surfactants, and inorganic salts. In Israel, research has demonstrated that the severity of powdery mildew on cucumber, grape, nectarine, mango, and rose can be reduced through foliar applications of phosphate and potassium salts [10,11]. Furthermore, foliar application of calcium chloride has been also reported to delay ripping and control mould disease in strawberries [12]. Application of sodium bicarbonate or calcium chloride significantly reduced the early blight incidence and severity [13]. They added that Calcium chloride proved higher efficacy for reducing both disease incidence and severity than that of sodium bicarbonate when applied either alone or combined with *Saccharomyces cerevisiae.* Calcium administered to the plant through the nutrient feed has been reported to be important for resistance to bacterial wilt resistance in tomato [14]. Moreover, it was found that $KHCO_3$ applications were effective in reducing the severity of powdery mildew on *E. japonica* and pumpkin [15,16]. Furthermore, Chiosan, in recent years, the importance of chito-saccharides as plant growth promoting and disease control agents has been emphasized [17,18]. Chitosan has been shown to induce defense responses in different plants [19,20]. Also, Chitosan oligomers was found to induce defense responses in grapevine leaves and a stimulation of chitinase and $\beta$-1,3-glucanase activities [18]. On the other hand, essential oils are promising alternative compounds which have an inhibitory activity on the growth of pathogens. It is possible that essential oils could be used in plant disease control as the main or as adjuvant antimicrobial compounds [21]. It is well established that some plants contain compounds able to inhibit the microbial growth [22]. These plant compounds can be of different structures and different mode of action

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when compared with antimicrobials conventionally used to control the microbial growth and survival [23].

The present work was aimed to evaluate the inhibitory effect of some plant resistance inducers against downy and powdery diseases incidence and severity of cantaloupe grown under plastic house conditions.

## II. MATERIALS AND METHODS

Evaluating the efficacy of different plant resistance inducers and/or bio-agents treatments against foliar diseases was carried out under plastic house conditions located at Nubaria region, Behiera Governorate, Egypt. The following treatments were applied as foliar spray:

1. Calcium chloride (20mM) + *S. cerevisiae* $10x10^{10}$ cfu/mL (10ml/L) + Chitosan (0.05mM)
2. Potassium bicarbonate (20mM) + Thyme oil ( 5ml/L)
3. Control (received only the fungicides approaches following the protective program recommended by Agriculture Research Centre, Protected Cultivation, Ministry of Agriculture and Reclaimed soil), Table (1).

**Table (1) Protective program against fungal diseases for growing cantaloupe in plastic houses and its financial cost ***

| No. | Time of Treatment | Disease | Fungicide | Concentration (ml & gm /100 liter) | Dose / plastic house (gm & ml) |
|---|---|---|---|---|---|
| 4 | After 7 days from previous spray | Downy mildew | Ridomil Gold | 200 gm | 200 gm |
| 5 | After 7 days from previous spray | powdery mildew | Tobaz –100 | 40 ml | 40 ml |
| 6 | After 7 days from previous spray | Downy & powdery mildews | Amistar- Top | 50 ml | 50 ml |
| 7 | After 10 days from previous spray | Downy mildew | Previcure –N | 250 ml | 250 ml |
| 8 | After 10 days from previous spray | Downy mildew | Aquagen-pro | 40 gm | 40 gm |
| 9 | After 7 days from previous spray | powdery mildew | Tobaz-100 | 40 ml | 40 gm |
| 10 | After 7 days from previous spray | powdery mildew | Sulpher | 300 gm | 300 gm |
| 11 | After 7 days from previous spray | powdery mildew | Copper Oxychloride | 300 gm | 300 gm |
| 12 | After 10 days from previous spray | Downy mildew | Previcure –N | 250 ml | 250 ml |
| 13 | After 7 days from previous spray | Downy & powdery mildews | Amistar- Top | 50 ml | 50 ml |

* The followed protective program is recommended by Agriculture Research Centre, Protected Cultivation, Ministry of Agriculture and Reclaimed soil

The experimental plastic house consists of 5 rows, each (0.9 x 60m, width x long) divided into 3 parts 20m long, and every part considered as one replicate. Five replicates were used for each particular treatment in complete randomized design. The growing vegetables were sprayed with proposed treatments 4 times with 15 days intervals after transplanting [24]. At all locations, the growing vegetables in the experimental plastic houses received only the recommended pesticides against harmful insects, *i.e.* aphids, trips, white fly, etc. as needed. Meanwhile, only the check control received traditional programs for controlling foliar diseases which recommended by the follow up committee of Protected Cultivation Administration Office, Ministry of Agriculture and Soil Reclamation. Monitoring and scouting of foliar diseases incidence, *i.e.* powdery and downy mildews of Cucumber, Pepper and cantaloupe & early and late blights of Tomato were recorded till the end of growing season. Percentages of disease incidence and severity were recorded at 60, 90 and 120 days of transplanting date. At the end of growing season the accumulated yield was calculated for each particular treatment in both experimental and control (Fungicide treatment).

***Disease assessment:***

- **Disease incidence:**

Percentage of each foliar disease incidence was recorded as the number of diseased plants relative to the number of growing plants for each treatment, then the average of disease incidence was calculated.

- **Disease severity:**

Percentage of each foliar disease severity was recorded as following equation:

$$D.S.\% = \frac{\Sigma ( n \times c )}{N} X100$$

Whereas: D.S. = Disease severity %
n = Number of infected leaves per
c = Category number
N= Total examined leaves

Disease severity scale from 0 to 4 according to [25] was followed, whereas: 0 = No leaf lesions; 1 = 25% or less; 2 = 26-50 %; 3 = 51-75 %; and 4 = 76-100% infected area of plant leaf.

***Statistical Analysis***

All experiments were set up in completely randomized design (CRD). The data collected from greenhouse experiment were analyzed by MSTAT-C program [26]. The

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means differences were compared by least significant difference test (LSD) at 5% level of significance. In plastic house experiments, one-way ANOVA was used to analyze differences between applied treatments. A general linear model option of the analysis system SAS [27] was used to perform the ANOVA. Duncan's multiple range test at $P \leq 0.05$ level was used for means separation [28].

## III. RESULTS AND DISCUSSION

Downy and powdery mildews diseases were recorded Cantaloupe grown in plastic house at Nubaria location,. Data in Table (2) show that both downy and powdery mildews incidence of cantaloupe were started to occur after 60 days from transplanting in significant difference at alternatives approaches comparing with fungicide control treatment. The downy and powdery mildews incidence were recorded as 0.0, 3.4, 7.8% and 0.0, 7.2, 10.2% at foliar treatments, [Calcium chloride + *S. cerevisiae* + Chitosan] and 0.0, 5.3, 9.6% and 0.0, 9.7, 12.7% at foliar treatment [Potassium bicarbonate + Thyme oil] comparing with 12.3, 26.3, 36.6 and 14.6, 27.4, 37.6% at fungicide treatment after 60, 90 and 120 days of transplanting, respectively. The applied foliar treatments could suppress the development of downy and powdery expression which recorded as 0.0, 0.5, 0.7% and 0.0, 0.4, 0.7% at foliar treatment of [Calcium chloride+ *S. cerevisiae* + Chitosan] and 0.0, 0.7, 0.9% and 0.0, 0.6, 0.9% at foliar treatment [Potassium bicarbonate + Thyme oil] comparing with 0.9, 1.3, 2.2% and 1.3, 2.3, 2.7% at fungicide treatment after 60, 90 and 120 days of transplanting, respectively (Table 3). Furthermore, reduction in downy and powdery mildews diseases incidence and severity of cantaloupe was also recorded (Fig. 1, 2). Reduction in downy and powdery mildews disease incidence was recorded as 100, 87.0, 87.6 & 100, 73.7, 72.8% at treatment of [Calcium chloride+ *S. cerevisiae* + Chitosan] and 100, 79.8, 73.7% & 100, 64.5, 66.2% at treatment of [Potassium bicarbonate + Thyme oil] after 60, 90 and 120 days of transplanting, respectively (Fig. 1).

**Table (2) Percentage of Downy and Powdery diseases incidence in response to application of plant inducers formula against foliar diseases of Cantaloupe grown in plastic houses under protected cultivation system (Nubaria location)**

| Treatment | Foliar diseases incidence (%) | | | | | |
|---|---|---|---|---|---|---|
| | Downy mildew | | | Powdery mildew | | |
| | Days of transplanting | | | Days of transplanting | | |
| | **60** | **90** | **120** | **60** | **90** | **120** |
| Calcium chloride + *S. cerevisiae* + Chitosan | 0.0 b | 3.4 c | 7.8 c | 0.0 b | 7.2 c | 10.2 c |
| Potassium bicarbonate + Thyme oil | 0.0 b | 5.3 b | 9.6 b | 0.0 b | 9.7 b | 12.7 b |
| Control (Fungicide pplication) | 12.3 a | 26.3 a | 36.6 a | 14.6 a | 27.4 a | 37.6 a |

Mean values within each column followed by the same letter are not significantly different ($P \leq 0.05$).

Also, [Calcium chloride+ *S. cerevisiae* + Chitosan] treatment could suppress both diseases development and expression in respective order by 100, 61.5, 68.1% and 100, 82.6, 74.0%. Meanwhile, reduction in diseases severity by 100, 46.1, 59.0% and 100, 73.9, 66.6% were recorded after 60, 90 and 120 days in respective order of plant growth period at treatment of [Potassium bicarbonate + Thyme oil] (Fig. 2).

**Table (3) Percentage of Downy and Powdery diseases severity in response to application of plant inducers formula against foliar diseases of Cantaloupe grown in plastic houses under protected cultivation system (Nubaria location)**

| Treatment | Foliar diseases severity (%) | | | | | |
|---|---|---|---|---|---|---|
| | Downy mildew | | | Powdery mildew | | |
| | Days of transplanting | | | Days of transplanting | | |
| | **60** | **90** | **120** | **60** | **90** | **120** |
| Calcium chloride + *S. cerevisiae* + Chitosan | 0.0 b | 0.5 c | 0.7 c | 0.0 b | 0.4 c | 0.7 c |
| Potassium bicarbonate + Thyme oil | 0.0 b | 0.7 b | 0.9 b | 0.0 b | 0.6 b | 0.9 b |
| Control (Fungicide application) | 0.9 a | 1.3 a | 2.2 a | 1.3 a | 2.3 a | 2.7 a |

Mean values within each column followed by the same letter are not significantly different ($P \leq 0.05$).

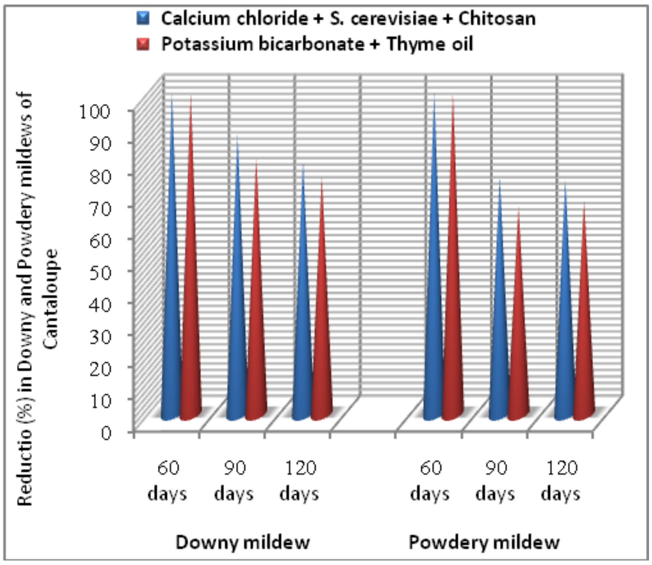


**Fig. (1) Reduction in Downy and powdery mildew diseases incidence in response to application of plant inducers formula against foliar diseases of Cantaloupe grown in plastic houses under protected cultivation system (Nubaria location)**

The accumulated yield of Cantaloupe, grown at Nubaria location, estimated as 0.502 and 0.492 ton per plastic house respectively at treatments of [Calcium chloride+ *S. cerevisiae* + Chitosan] and [Potassium bicarbonate + Thyme oil] with an increase of 14.6 and 12.3 over fungicide treatment (Table 4 and Fig. 3). The accumulated yield and its increase in vegetables response to applied alternatives approaches comparing with fungicide application was calculated and recorded. The applied fungicide alternatives approaches, [Calcium chloride+ *S. cerevisiae* + Chitosan]

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and [Potassium bicarbonate + Thyme oil] could reduce both foliar diseases incidence and suppress their development which reflected on increase the obtained yield of treated cantaloupe plants. The use of plant resistance inducers in combination with bio-agents was subjected to evaluation in many reports. In this regards, an interesting alternative to fungicide application for plant disease control involves the use of some organic and inorganic salts with antimicrobial properties generally used in food processing and preservation. Selected organic and inorganic salts are active antimicrobial agents and have been widely used in the food industry. Many of these salts are effective against a range of microorganisms; most of them have low mammalian toxicity and therefore have potential for postharvest disease control. Salt treatments can inhibit plant pathogens or suppress mycotoxin production [28,30].

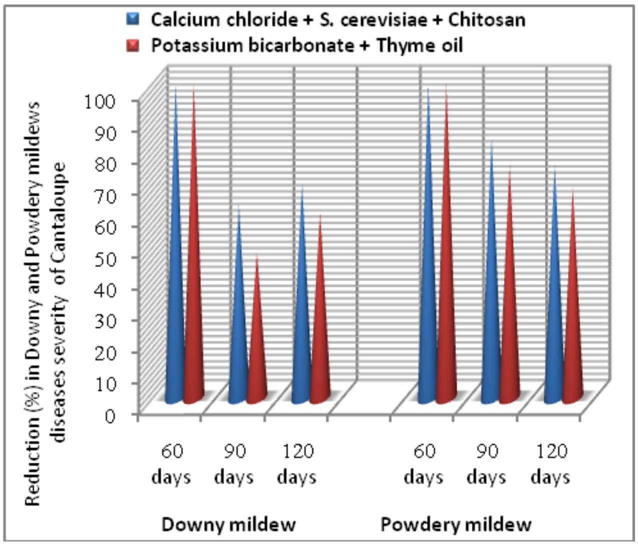


**Fig. (2) Reduction in Downy and powdery mildew diseases severity in response to application of plant inducers formula against foliar diseases of Cantaloupe grown in plastic houses under protected cultivation system (Nubaria location)**

Also, [31] found that from field experiments, that spraying cantaloupe plants three times with fungicides in alternation with another three sprays with any of calcium chloride or salicylic acid resulted in significant reduction in the disease severity with significant increase in the fruit yield when compared with unsprayed (check) plants. Similar reports conducted the efficacy of chemical inducers application individual or combined with bioagents against plant pathogens were cited in literature. The use of sodium bicarbonate alone to control postharvest decays of fruit has its limitations [32], but it can be combined with other alternative treatments to synthetic fungicides, resulting in the control that is superior to individual treatments alone. For example, sodium bicarbonate was successfully used in combination with bacterial and yeasts biocontrol agents to enhance control of postharvest decays on citrus, pome, and stone fruits [33,34]. These reports are clearly demonstrated in the present study and show that the application of *S. cerevisiae* enhanced the control of foliar cantaloupe diseases when combined with either calcium chloride spray. Many researchers have shown that calcium plays an important role in the inhibition of postharvest decay of fruits [35,36], and in enhancing the efficacy of postharvest bio-control agents [37,38].

**Table (4) Obtained yield of Cantaloupe in response to foliar application of plant inducers formula in plastic houses under protected cultivation system (Nubaria location)**

| Treatment | Average Yield Kg/row | Yield Ton/plastic house | Increase % |
|---|---|---|---|
| Calcium chloride + *S. cerevisiae* + Chitosan | 100.4 a | 0,502 a | 14.6 |
| Potassium bicarbonate + Thyme oil | 98.4 b | 0,492 b | 12.3 |
| Control (Fungicide application) | 87.6 c | 0,438 c | - |

Mean values within each column followed by the same letter are not significantly different ($P \leq 0.05$).

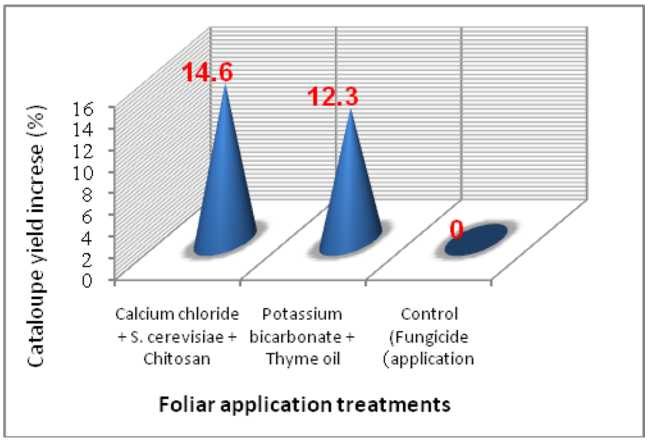


**Fig. (3) Yield increase in response to application of plant inducers formula against foliar diseases of Cantaloupe grown in plastic houses under protected cultivation system (Nubaria location)**

As for Chiosan, in recent years, the importance of chitosaccharides as plant growth promoting and disease control agents has been emphasized [17,18]. Chitosan oligomers was found to induce defense responses in grapevine leaves, as evidenced by an accumulation of stilbene phytoalexins, *trans*- and *cis*-resveratrol, ε-viniferins, and piceids, and a stimulation of chitinase and *β*-1,3-glucanase activities [17]. They added that the combination of Chitosan and $CuSO_4$ increased phytoalexin production. This elicitor capacity of Chitosan and/or $CuSO_4$ appeared to be associated with an induced protection of grapevine leaves against gray mold and downy mildew diseases. Also, Chitosan enhanced the accumulation of pathogenesis related-proteins such as ss-1,3-glucanase, chninase and PR14 in treated and upper untreated tomato leaves [39]. Moreover, several workers suggested two different mechanisms of chitosan molecule and target microorganism interaction: the first is the adsorption of chitosans to cell walls leading to the cell wall covering, membrane disruption and cell leakage; the second is the penetration of chitosans into living cells leading to the inhibition of various enzymes

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and interference with the synthesis of mRNA and proteins [40,41]. Essential oils as natural alternatives that are user friendly and demonstrate low toxicity to humans are desirable to be tested either alone or in combination in the present work. Thyme oil applied in combination showed effective reduction in foliar diseases incidence. In this regards, several investigators reported the antifungal effect of essential oils. Thyme and Egyptian geranium oils are considered antimycotic natural compounds may be useful for inhibition of mold fungi on wood in service or during storage of building materials [42]. Moreover, [43] had the first report on the use of Thymol for controlling a plant disease under field conditions, which indicated that this compound pro-vided effective control of bacterial wilt on susceptible tomato cultivars. Also, Thymol has been reported to have fungicidal activities and fumigation with thymol has been used for control of postharvest fungal diseases [44, 45]. Modes of action of the antibacterial property of thymol appeared to include disruption of bacterial cell membrane integrity by altering protein reactions [46,47]. The obtained results reported here, foliar application of plant resistance inducers could become effective components of an integrated disease management system for cantaloupe. It is also suggested that such application trials with downy and powdery mildews diseases on other vegetable crops be conducted.

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