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EFFECT OF SOME MICRONUTRIENTS AND THE ANTIOXIDANT SALICYLIC ACID ON SUPPRESSING THE INFECTION WITH STRAWBERRY POWDERY MILDEW DISEASE

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ABSTRACT

Spraying strawberry plants with three micronutrients, *i.e.*, ferrous (Fe) , manganese (Mn) and zinc (Zn) , singly or in different combinations, and in combination with the inducing systemic acquired resistance (ISAR) salicylic acid significantly reduced the severity of infection with powdery mildew disease , caused by *Sphaerotheca macularis* ,and increased fruit yield compared with the control. In addition, spraying ISAR salicylic acid alone was more effective in reducing the disease than spraying micronutrients alone. Furthermore, spraying the plants with ISAR salicylic acid three times in combination with at least two of the tested micronutrients was a more effective treatment in this regard, where no apparent symptoms of the disease were observed and also resulted in the highest fruit yield, total soluble solids (T.S.S.) and fruit firmness.

Key words: *micronutrients, powdery mildew , salicylic acid , strawberry, systemic acquired resistance ,total soluble solids and fruit firmness.*

1. INTRODUCTION

Strawberry (*Fragaria* X *ananassa* Duch.) is one of the most

important nontraditional vegetable crops in Egypt, for which the demand has increased for local consumption and exportation. Also, it is one of the most favorable fruits either for fresh consumption or food processing . The cultivated area of strawberry (both Fresh and Frigo plantations) reached more than 6300 feddan during 2002 / 2003 growing season with an average of 18 ton / feddan for fresh plantation and 12 ton for Frigo plantation (Higher Committee of Strawberry , Min. of Agric., 2003).

Under Egyptian conditions, strawberry plants are vulnerable to infection with many destructive diseases, *i.e.* wilt and root and crown-rots (Fahim *et al.*, 1994 a and El-Kolaly, 2002), fruit-rots (Khafagy, 1982 ; and Abada *et al.*, 2002) and leaf blight and spot (Tadrus, 1999 and Attia, 2004), in addition to minor infection with viral and bacterial diseases.However, powdery mildew caused by *Sphaerotheca macularis* is one of the most destructive diseases due to its harmful effect on the foliage growth and the quantity and quality of fruits.

Chemical control mostly causes environmental pollution, highly affects the growth of the host plants, leads to great disturbance of the natural biological balance and the most serious result in the great increase in the accumulation of the toxic substances in the human food chain. Therefore, this research aimed to study the possibility of suppressing the strawberry powdery mildew disease infection with spraying a combination of some safe agrochemicals such as micronutrients and inducing systemic acquired resistance (ISAR) salicylic acid under greenhouse conditions.

2. MATERIALS AND METHODS

Pot experiment was used to study the effect of spraying ISAR in combination with some micronutrients on suppressing the infection with strawberry powdery mildew disease caused by *Sphaerotheca macularis*.

It is worth to mention that field observations revealed that Silva cv. is the most susceptible to this disease, therefore it was used throughout this experiment.

Virgin sandy soil was thoroughly washed twice daily for five days with tap water to remove the excess salts (strawberry plants are sensitive to salinity (Ulrich *et al.*, 1980). Plastic pots (No.25) were

then filled with the washed sandy soil. Each pot received super phosphate, ammonium and potassium sulfate fertilizers at the rate of 2.0 g from each fertilizer , then irrigated. Two frigo transplants of Silva cv. (previously soaked in the fungicide Rizolex-T at the rate of 0.1 % for half hour just before transplanting), were transplanted in each pot. One month after transplanting, the tested micronutrients, *i.e.* ferric sulfate (Fe) , magnesium sulfate (Mn) and Zinc sulfate (Zn) were sprayed one month after transplanting, either singly or in different combinations, four times (one month interval), at the rate of 0.1 %. Meanwhile, the induced systemic acquired resistance salicylic acid (ISAR) was sprayed at the rate of 0.05%(dissolved in acetone just before spraying), three times, *i.e.* 45, 60 and 75 days after transplanting. Also, the recommended amounts of the other fertilizers were added to the growing plants every two weeks three months after transplanting. Naturally infected leaves of Silva cv. were collected from strawberry fields and used for inoculation . In this respect, the collected infected leaves were shaken up on the growing plants, previously sprayed with distilled water. Uninoculated plants as well as unsprayed plants with the micronutrients and the ISAR served as controls. Four replicate pots were used for each treatment.

The produced fruit yield of each treatment was harvested periodically and weighed (when it was necessary) and the averages were recorded . Also, five fruits were taken randomly from each harvesting to determine their total soluble solids (T.S.S.) using a hand refractometer and firmness using pressure tester (pound / inch ^{2}) and the averages were recorded.

2.1.Disease assessment

The inoculated plants were examined weekly for disease symptoms and disease severity was assessed using the devised scale by Horsfall and Barratt (1945) each month (three times), then the averages were calculated.

2.2.Statistical analysis

Data were statistically analyzed using the split plot design (Snedecor and Cochran , 1967). The averages were compared at 5 % level of probability by the L.S.D. test.

3. RESULTS

3.1. Effect on disease severity and fruit yield

Data presented in Table (1) show that spraying strawberry plants (cv. Silva) with the induced systemic acquired resistance ISAR salicylic acid in combination with some micronutrients generally resulted in a significant reduction in the severity of powdery mildew disease with significant increase in the fruit yield compared with the control. Moreover, spraying the ISAR salicylic acid only was more efficient than spraying micronutrients in this regard. Furthermore, no apparent infection with the disease was observed when the plants were sprayed three times with ISAR in combination with any of Fe + Mn, Fe + Zn , Mn + Zn and Fe + Mn + Zn . However, low disease severity was found when the plants were sprayed with the ISAR salicylic acid three times in combination with any of Fe , Mn, and Zn , being 1.3 ,1.0 and 1.0 % on the average, respectively. The control of the previous treatments recorded 8.0 % disease severity. The average of disease severity on unsprayed plants with the ISAR or micronutrients was 42.0 % . Meanwhile the averages for sprays 1,2 and 3 times were 22.0 , 10.0 and 8.0 % , respectively .

The combination between the tested micronutrients and the ISAR salicylic acid was greatly reflected on the fruit yield, which recorded significant increase in the produced fruit yield in the case of sprayed plants compared with unsprayed ones.

3.2. Effect on the total soluble solids (T.S.S.) and firmness of the fruits

Data (Table 2) show the effect of spraying strawberry plants with the ISAR salicylic acid in combination with some micronutrients on T.S.S. and firmness of the produced fruits. Results revealed that there were , to some extent , low differences in the estimated T.S.S. due to the using of the micronutrients and ISAR compared with the control. However, spraying the three micronutrients in combination with the IASR salicylic acid produced fruits of high T.S.S. , being 10.8 % compared with spraying any micronutrient alone or in combination with ISAR. On the other hand, unsprayed plants produced fruits poor in T.S.S., being 8.1 %.

Table (1): Effect of spraying strawberry plants (cv. Silva) with three micronutrients in combination with the ISAR salicylic acid on infection with powdery mildew and fruit yield.

Micronutrients	% Disease severity after spraying ISAR (1-3 times)				Mean	Average fruit yield(g)/plant after spraying ISAR (1-3 times)				Mean
	0.0	One	Two	Three		0.0	One	Two	Three	
Fe	35.1	10.0	4.2	1.3	12.7	222	228	230	233	228.3
Mn	34.0	11.3	4.0	1.0	12.6	220	228	231	233	288.0
Zn	34.0	11.0	4.0	1.0	12.5	219	228	231	233	227.8
Fe + Mn	33.0	9.8	3.6	0.0	11.9	228	232	234	236	232.5
Fe + Zn	32.1	9.8	3.6	0.0	11.4	229	233	234	237	233.3
Mn + Zn	31.2	9.6	3.2	0.0	11.0	231	235	237	239	235.5
Fe + Mn + Zn	29.1	6.0	1.7	0.0	9.2	252	256	258	258	256.0
Control	42.0	22.0	10.0	8.0	20.5	196	200	201	203	200.0
Mean	33.3	9.6	3.5	0.5	-----	224.6	230.0	232.0	234.0	------
L.S.D. at 5 % for : Micronutrients (M) =					1.2					4.2
No.of sprays (S) =					2.4					2.9
M x S =					3.3					3.8

Table (2): Effect of spraying strawberry plants (cv. Silva) with three micronutrients in combination with the ISAR salicylic acid on T.S.S. and firmness of the produced healthy fruits.

Micronutrients	%, T.S.S. of fruits after spraying ISAR (1-3 times)				Mean	Firmness of fruits after spraying ISAR (1-3times)				Mean
	0.0	One	Two	Three		0.0	One	Two	Three	
Fe	8.4	8.4	8.4	8.4	8.4	2.8	2.8	2.9	3.0	2.9
Mn	8.0	8.1	8.2	8.2	8.2	2.8	2.9	2.9	3.0	2.9
Zn	8.0	8.1	8.3	8.3	8.2	3.0	3.1	3.1	3.2	3.1
Fe + Mn	4.0	9.2	9.2	9.2	9.2	3.1	3.2	3.2	3.3	3.2
Fe + Zn	9.2	9.3	9.3	9.4	9.3	3.1	3.2	3.2	3.3	3.2
Mn + Zn	9.4	9.5	9.5	9.5	9.5	3.1	3.2	3.2	3.3	3.2
Fe + Mn + Zn	10.6	10.8	10.8	10.8	10.8	3.7	3.7	3.7	3.8	3.7
Control	8.1	8.4	8.5	8.5	8.4	2.1	2.3	2.4	2.4	2.3
Mean	8.9	9.1	9.1	9.1	------	3.0	3.1	3.1	3.2	------
L.S.D. at o.o5 for : Micronutrients (M) =					0.6					0.5
No.of sprays (S) =					n.s.					n.s.
M x S =					0.8					1.1

ISAR salicylic acid in combination with some micronutrients caused significant increase in fruit firmness compared with the control. On the other hand, no significant differences were detected due to the effect of the number of sprays. In addition, spraying the three micronutrients together in combination with the IASR salicylic acid was the superior treatment in this regard recording 3.8 % fruit firmness. Meanwhile, spraying each of Fe, Mn, singly or in combination with the ISAR salicylic acid recorded the lowest fruit firmness values, being 2.8, 2.9 % on the average, respectively without significant differences.

4. DISCUSSION

The current strategy of controlling pests of vegetables and fruits depends on using alternative methods of disease control rather than pesticides and fungicides and/or using their chemicals at the first periods of plant growth prior to fruit formation. The infection with strawberry powdery mildew occurs mainly at the time of fruit production and on the formed fruits, hence this work aimed to using ISAR (safe chemical) in combination with some micronutrients for management this disease.

The obtained data indicated that spraying strawberry plants with salicylic acid for the induction of systemic resistance in combination with some micronutrients, *i.e* Fe, Mn and Zn, sigly or in combinations, resulted in a significant reduction in powdery mildew disease infection with considerable increase in fruit yield and fruit T.S.S. and firmness compared with the control. In this respect, spraying the ISAR in combination with any of Fe + Mn , Fe + Zn , Mn + Zn and Fe + Mn + Zn resulted in a complete suppression to the disease with high fruit yield, T.S.S. and fruit firmness.

The systemic acquired resistance is an important component of plant's defense against diseases, where initial infection provides systemic resistance to subsequent infection by a variety of bacterial , fungal and viral pathogens (Gaffieny *et al.*, 1993).

The reduction in powdery mildew disease herein supports the hypothesis that induced acquired resistance induced by restricted infection is not due to a specific component of the pathogen, but rather to gradual appearance and gradual persistence of a level of metabolic perturbation leading to stress on the host . Dean and Kuc

(1985) and Doubrava *et al.*(1988) indicated that induced acquired resistance is persistent and generally is pathogen nonspecific. In addition, systemic acquired resistance can be induced by simple substances as well as by biotic agents. On the other hand, Lancke (1981) found that unlike elicitors of phytoalexins accumulation, which are elicited at the site of application may be responsible for localized protection and induces systemic acquired resistance that sensitizes the plant respones rapidly after infection. These responses induced phytoalexins accumulation and lignification (Dean and Kuc ,1985 and Kuc and Rush, 1985) and induce or enhance activities of chitinase and β glucanse (Metreux and Boller, 1986). Furthermore, Kessmann *et al.* (1994) mentioned that the mechanism of systemic acquired resistance is apparently multificated, likely resulting in stable, broad spectrum disease control and they could be used preventatively to bolster general plant health, resulting in long lasting protection.

Chen, *et al.* (1993) and Chondra *et al.* (2001) cloned and sequenced the salicylic acid binding protein which exhibited catalase activity. They also reported that the action of salicylic acid in systemic acquired resistance is obtained by elevated amounts of H_2O_2 by inhibiting the catalase activity. Apart from this, it has also been reported that salicylic acid induces many proteins similar in nature to the pathogenesis related proteins (Linthorst, 1991).

The use of salicylic acid to induce systemic acquired resistance was previously used to minimize the infection with many diseases (Malamy and Kessing, 1992 Dekker, 1996; Sticker *et al.* 1997; Ibrahim, 1998; Bhatt *et al.*, 1999; Abou Taleb, 2001; Pradeep and Jambhale, 2002 and Attia ,2004).

It seems from the present results that all the three tested micronutrients have a role in increasing the resistance to the disease, which play a role in the enzymes formation and their activity in the plant during their growth as many enzymes are of organic complex structure (Dekock *et al.*,1959; Salama, 1990 and Fahim *et al.*,1994 b). Moreover, Yaroshenko (1962) reported that micronutrients affected oxidative-reduction system in the plant.

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تأثير ثلاثة عناصر صغرى وحامض السالسيليك المضاد للأكسدة على تثبيط الإصابة بمرض البياض الدقيقي في الفراولة

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ملخـــص

أدى رش نباتات الفراولة بثلاثة مغذيات صغـــرى (الحديـد ، الزنـك ، المنجنيز ، كل على حده أو مع بعضها) مع حـــامض السالســيلك (المحـدث للمقاومة الجهازية المكتسبة) إلى إحداث انخفاض معنوي لشدة الإصابة بمـــرض البياض الدقيقي المتسبب عن الفطر سفيروسيكا ماكيولاريز مع زيادة معنوية فـي محصول الثمار بالنسبة لنباتات المقارنة. بالإضافة إلى ذلك فقـد كـان الـرش بحامض السالسيلك هو الأكثر فعالية في خفض الإصابة بـــالمرض عـن الـرش بالعناصر الصغرى وحدها. علاوة علي ذلك فقد كـان رش النباتـات بحـامض السالسيلك ثلاث مرات مع استخدام عنصرين من المغذيات الصغرى، علي الأقل، هو الأفضل في هذا الصدد، حيث لم تشاهد أي أعراض ظاهرية للمرض مع إنتاج أعلي محصول لثمار الفراولة من حيث المواد الصلبة الذائبة والصلابة العالية.

المجلـة العلمية- لكلية الزراعة-جامعة القاهرة - المجلد (٥٥) العدد الثـالـث (يوليو ٢٠٠٤): ٤٧٥-٤٨٦.