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Ability of Mineral Salts and Some Fungicides to Suppress Apple Powdery Mildew Caused by the Fungus *Podosphaera leucotricha*

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ABSTRACT

A leaf bioassay was done *in vitro* to determine the effects of different mineral salts on the infection of apple powdery mildew. Calcium chloride, cupper sulfate and calcium carbonate led to more than 50% significant reduction in disease severity % and were more effective than ammonium phosphate and potassium dihydrogen phosphate. Golden Delicious and Royal Gala were found susceptible to the disease thus were subjected for post-infection treatment with some systemic fungicides or calcium chloride under field conditions. One week after the foliar spray, Triforine and Triadimenol were more effective than Carbendazim and calcium chloride in reducing the disease amount. Their reduction in disease progress was continued after one month from the day of the foliar spray. Calcium chloride was more effective at an application rate of 10 g L^{-1} than at 1 g L^{-1}. Calcium chloride treatment did not give long-term efficacy especially in Royal Gala. Therefore, the use of salts as an anti-resistance strategy should be used in a rotation with chemical fungicides or combined with other control measures.

Key words: Cultivars, fungicides, management, minerals

INTRODUCTION

Powdery mildew is one of the most serious common diseases of apples (Wurms and Chee, 2011). Powdery mildew occurs in all apple-growing areas around the world especially in semiarid regions including Jordan (Qasem and Abu-Blan, 1986; Abdel-Rahman, 1996). The disease caused by the pathogenic fungus; *Podosphaera leucotricha* (Ell. and Ev.) Salm. Economic disease losses depend on the susceptibility of cultivars, environmental conditions and disease management strategies (Turechek *et al*., 2004). The disease causes a considerable loss because it reduces the photosynthetic area of leaves in young non-bearing plants, which can severely stunt the growth of seedlings and can greatly reduce yield (50-100% in the absence of control) in mature plants. Infections on fruit cause russeting resulting in bad fruit quality and value (Yoder, 1992).

Management of powdery mildew is usually based on prevention or reduction of primary infections of powdery mildew. Chemical control is commonly used for apple protection from powdery mildew. Fungicide application reduces primary infections and prevents secondary infections of leaves and buds (Drimal *et al*., 2007; Gupta and Sharma, 2005). The effects of different salts were previously investigated on many diseases attacking apple and other plant types (Conway *et al*., 2004; Kanto *et al*., 2004; Creemers *et al*., 2008). However, the practical use of salts as an environmentally sound fungicide in integrated farming systems is still confined. Therefore, this study was conducted to determine the effects of some salts *in vitro* and some systemic fungicides on controlling powdery mildew of apple under field conditions.

MATERIALS AND METHODS

***In vitro* leaf bioassay:** *In vitro* leaf bioassay was performed as follow: apple leaves (5 cm-in-diameter) showing about 30% disease severity of powdery mildew were individually picked from diseased apple trees cv. Golden Delicious and dipped in 100 mL of different mineral salts (technical grades); ammonium phosphate ($(NH_4)_3PO_4$), potassium dihydrogen phosphate (KH_2PO_4), calcium carbonate ($CaCO_3$), calcium chloride ($CaCl_2$) and cupper sulfate ($CuSO_4$) at 1 and 10 g L^{-1} concentrations. A control was added by dipping the leaves in distilled water for 1 min. Each leaf was placed into a test tube over a cotton ball moistened with 10 mL distilled water. All tubes were closed with screw caps and incubated in a full-controlled growth chamber at 24°C with a 12 h photoperiod under fluorescent light (3000 lux day^{-1}) for one week. All treatments and control were arranged in a Completely Randomized Design (CRD). Each treatment was replicated five times and whole experiment was repeated twice. Disease severity of powdery mildew was assessed using the score chart of 0 to 5 scale; 0: no infection, 1: 0-10, 2: 10.1-15, 3: 15.1-25, 4: 25.1-50% and 5: more than 50% leaf areas covered with mildew growth (Jamadar and Desai, 1997; Anand *et al.*, 2008). Disease reduction % was then calculated as the decrease of the amount of disease (disease severity %) due to a treatment divided on the amount of disease in the untreated control.

Field characteristics and disease assessment: A field study was conducted in a commercial apple orchard during 2010 apple growing season at Al-Shoubak area of Jordan, situated approximately 260 km south of Amman. The orchard is planted with Golden Delicious and Royal Gala apple trees (5-years-old and 2.5 m height) grafted on MM111 rootstocks and planted into 2 m-spaced rows. Spacing between trees was 2 to 4 m (about 1250 trees ha^{-1}). The disease caused by *Podosphaera leucotricha* was evident in the orchard in the previous years. Methods of fertilization, irrigation and other cultural practices for this crop were as recommended to commercial growers (Extension Service, Ministry of Agriculture, Jordan). The mean daily atmospheric temperature ranged from 11.9-33°C and relative humidity ranged from 50-59% during the experiment period in 2011 apple growing season. Four treatments were arranged in a randomized complete block design. Plots were five apple trees replicated two times per cultivar. The treatments included an untreated control (sprayed with only water), the systemic fungicides Triadimenol (Bayfidan®, 250 Emulsifiable Concentrate), Triforine (Saprol®, 190 Emulsifiable Concentrate) and Carbendazim (Mycozim®, 50% Soluble Concentrate) belonging to different chemical groups; Triazoles, Piperazines or Benzimidizoles group, respectively were selected and used at their low and high commercial application rates plus the use of calcium chloride salt at 1 and 10 g L^{-1} concentrations. A foliar spray was performed 2 weeks after bud break at the morning using a knapsack sprayer to cover whole tree canopy with the fungicide or salt solution. Disease reduction % was assessed as previously described and recorded one week and one month after treatment.

Statistical analysis: Data were analyzed statistically using General Linear Model (GLM) procedure (SPSS software version 11.5; SPSS Inc., Chicago, USA). Least significance difference (LSD) test and t-test were used for mean separation at the 0.05 probability level (Steel *et al.*, 1997).

RESULTS AND DISCUSSION

Powdery mildew of apple caused by the fungus *Podosphaera leucotricha* was found to occur in apple orchards at Al-Shoubak area of Jordan. Both Golden Delicious and Royal Gala apple cultivars were found susceptible to powdery mildew caused by *Podosphaera leucotricha*. After one week from the *in vitro* leaf incubation, the disease continued its development from 30 to 74.3% in the control (Table 1). Among the tested mineral salts, calcium chloride (at 1 and 10 g L^{-1}) and cupper sulfate at 10 g L^{-1} led to the highest significant reductions in DS% values and caused more than 50% reduction (77.6, 78.0 and 64.1%, respectively). Ammonium phosphate and potassium dihydrogen phosphate at 1 g L^{-1} did not cause a significant reduction in DS% and their effect was similar to the control. Calcium carbonate was only effective at 10 g L^{-1} in causing 50.7% reduction (Table 1).

One week after the foliar spray, Triforine and Triadimenol were more effective than Carbendazim and calcium chloride in reducing the disease (Fig. 1). The higher application rates gave more control of the disease than the lower rates. Calcium chloride caused a significant reduction in the disease amount and was more effective at 10 g L^{-1} than at 1 g L^{-1} application rates. The reduction in disease progress was slightly continued after one month from the day of the foliar spray except in the calcium chloride treatment of Royal Gala where the % of disease reduction was similar to that of the untreated control (Fig. 1).

Powdery mildew of apple was found to occur in Al-Shoubak area of Jordan. Results of a previous study revealed the occurrence of 26 powdery mildew fungal species parasitizing 156 host plant species in Jordan (Qasem and Abu-Blan, 1986). Both Golden Delicious and Royal Gala apple cultivars were found susceptible to powdery mildew caused by *Podosphaera leucotricha*. Among tested apple cultivars, Golden Delicious (Jeger and Butt, 1986; Marine *et al*., 2010) and Royal Gala (Wurms and Chee, 2011) were previously reported as susceptible cultivars.

Chemical treatments represented by three systemic chemical fungicides Triadimenol, Triforine and Carbendazim and a mineral salt (calcium chloride) were applied against the early infection of apple powdery mildew. More pronounced effect of the chemical treatment was observed after one week then gradually reduced. The efficacy of the systemic fungicides were gradually reduced but extended to one month after treatment. The long-term efficacy could reduce the number of

Table 1: Effects of different mineral salts on powdery mildew of apple *in vitro*

Salts	Application rate (g L^{-1})	Disease severity (%)	Disease reduction (%)
Ammonium phosphate	1	63.3[1]ab[2]	14.8[bc]
	10	56.7[bc]	23.7[bc]
Calcium carbonate	1	51.7[bc]	30.5[bc]
	10	36.7[de]	50.7[a]*
Potassium dihydrogen phosphate	1	45.0[cd]	39.4[b]
	10	73.3[a]	1.3[bc]
Calcium chloride	1	16.7[f]	77.6[a]*
	10	16.0[f]	78.0[a]*
Cupper sulfate	1	46.7[cd]	37.2[ab]
	10	26.7[ef]	64.1[a]*
Control (untreated)	-	74.3[a]	0.0[d]

[1] Average of 5 replicates, [2] Means within columns followed by the same letters are not significantly different at 0.05 probability level using LSD test, * More than 50% control

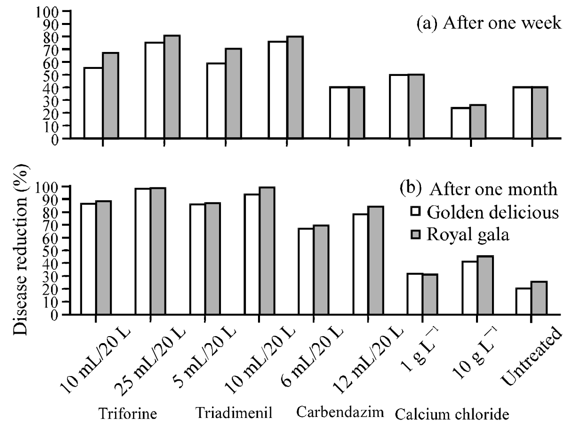

Fig. 1(a-b): Effects of fungicides and calcium chloride on powdery mildew disease as disease reduction % (a) After one week: LSD = 11.1 and (b) After one month: LSD = 9.5 from foliar application in two apple cultivars, golden delicious and Royal Gala, under field conditions

fungicide applications and could affect positively apple fruit yield. Chemical treatment of powdery mildew resulted in an increase of apple fruit yield by 37.3-13.9% while treatment with an ecological agent increased fruit yield by 25.9-12.2% in comparison to untreated (Drimal *et al.*, 2007). Also, less frequent post-infection applications of fungicides would reduce the cost of disease control in an economically sound manner (Pearson *et al.*, 1978). Triadimenol and Triforine were among the best fungicides that had controlled powdery mildew of roses (Wojdyla, 1994). Generally, the high application rates of the fungicides are more efficient than the low rates. There was a gradual increase in the severity and incidence of powdery mildew infection with time after spraying since the residual fungicide effect is usually decreasing with time. In another study (Lesnik and Bercic, 2001) when fungicides at 100, 85, 65 and 50% dosages were applied the rate of mildew infection on apple shoots significantly increased in all three trial years due to the reduction of fungicide dosages. Triforine was found to provide 89-98% control one day after inoculation followed by 59-94 and 68-83% control after two and three days, respectively for controlling cedar apple rust caused by *Gynmosporangium juniperi-virginianae* (Pearson *et al.*, 1978).

Mineral salts, detergents, anhydrous milk fat could be used an important component of an IPM strategy for control of powdery mildew on apple (Reuveni *et al.*, 1996, 1998; Creemers *et al.*, 2007; Sholberg and Boule, 2009; Wurms and Chee, 2011). For all the salts evaluated, calcium chloride and cupper sulfate showed high efficacy against powdery mildew on apple *in vitro*. Therefore, they can be considered as an ideal product for low infection risk moments. Creemers *et al.* (2007) considered potassium bicarbonate ($K(HCO_3)_2$) effective in controlling apple powdery mildew but less reliable during high infection risk and recommended to use $K(HCO_3)_2$ in combination with a chemical fungicide as the efficacy of $K(HCO_3)_2$ fluctuates. Calcium carbonate was moderately effective on powdery mildew since caused about 50% reduction and was less effective than calcium chloride and cupper sulfate. Similar results were previously reported. Direct spray of sodium bicarbonate at a concentration of 2,000 ppm inhibited 80-100% of conidial germination of powdery mildew of cucumber caused by *Sphaerotheca fuliginea* but the reduction percentage being only

38% (Homma *et al.*, 1981). Therefore, there is inconsistency about the robustness of salt treatments compared to chemical treatments makes salts less popular to be used in conventional disease management.

Combining chemical fungicides which is the best anti-resistance strategy, is not always possible or recommended when there is a limited number of available chemical fungicides or a reduction in fungicide use could be requested (Creemers *et al.*, 2007).

Potassium dihydrogen phosphate and ammonium phosphate were less effective than the non-phosphate salts and failed to cause more than 50% disease reduction. Previous studies have shown that potassium monohydrogen phosphate sprayed alone, as a tank mix or in alternation with fungicides have been successful in the control of powdery mildew in apples and some other hosts of powdery mildew (Napier and Oosthuyse, 1999). But since the method of salts application depends largely on the disease pressure and the crop to be sprayed. The benefits of incorporating salts into a spray program include good disease control, reduced fungicide applications (cost-effective), reduced fungicide residues on food crops, less fungicide resistance pressure, promotion of IPM and enhanced plant nutrition (Reuveni *et al.*, 1998). The fluctuation in the efficacy of calcium chloride on controlling powdery mildew level may be related to the heterogeneity of environmental conditions prevailed in the orchard. Similarly, the efficacy of another salt ($K(HCO_3)_2$) was reported to fluctuate under open field conditions (Creemers *et al.*, 2007).

In a conclusion, the use of salts as an anti-resistance strategy should be combined with or used in a rotation with chemical fungicides or combined with biological, mechanical and cultural control measures.

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