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Beneficial and Deleterious Effects of Phosphate Salts on Flax Plants Infected with Powdery Mildew

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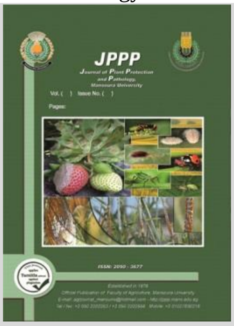

ABSTRACT

Four sprays of 25mM, 50mM, and 100mM solutions of NaH_2PO_4, Na_2HPO_4, KH_2PO_4, K_2HPO_4, $NH_4H_2PO_4$, and $(NH_4)_2HPO_4$ were used to compare their effects in controlling powdery mildew of flax. In this study, it was found that sodium monohydrogen phosphate Na_2HPO_4 was the only effective salt in suppressing the disease as it reduced powdery mildew severity by 44.75% when it was applied at the concentration of 50mM, other salts such as potassium monohydrogen phosphate significantly increased disease severity and they also caused significant reduction in seed and straw yield. The results of the present study indicated that concentration plays a decisive role in determining the effectiveness of phosphate salts in suppressing flax powdery mildew. Therefore, phosphate salts will be reevaluated in a future study by using lower concentrations as these lower concentrations could be effective in controlling the disease .

Keywords: Phosphate salts, Powdery mildew, Flax, Foliar spray.

INTRODUCTION

Flax (*Linum usitatissimum* L.) 2n=30, is an important source of bast fibers (phloem fibers) and seed oil worldwide and it is annual crop. Flax, commonly known as linseed (flax seed) or fiber flax depending on its main use, oil seed or fiber crop (Sertse *et al*., 2019). Flax is affiliated with the genus *Linum* of the family Linaceae. The name *Linum* came from lin or thread and the species name *usitatissimum* means the most beneficial (Dhirhi *et al*., 2017).

Flax seed contains α-linolenic acid, which is an essential omega-3 fatty acid, amino acids, vitamins, trace elements, lignans, dietry fiber and linseed gum, which has high dietotherapy and health care functions (Zhang *et al*., 2020). The oil flax is also important in the manufacture of soap, paints and printers ink (El- Nagdy *et al*., 2010)

Bast fibers are prevalently used in paper, packaging, and textile industries. The high biomass and easy availability of bast fibers make them suitable for industerial uses, like cellulose production, for commercial uses, like textile production, and for livestock production, as a nutritional feed stock. Flax produces fine fibers called linen, which are popular and in demand globally, particularly in the textile industry (Majumder *et al*., 2020).

Powdery mildew (PM), caused by the obligate biotrophic ascomycete *oidium lini* skoric, is a widely distributed and devastating disease of flax (*Linum usitatissimum* L.). The disease appears on all the plant parts (the stem, the leaves, the flowers and the capsules) as a white powdery mass of mycelia that starts as small spots on the leaves and spreads in a rapid way to cover all the surface of the plant. Flax plant, which is early infected may be exposed to severe defoliation and reduced yield and quality (Ashry and Mohamed, 2011). However, disease intensity differs from season to season depending on the location, fertilization, weather conditions, and cultivars (Aly *et al*., 2013). PM occurs every year in all flax production areas in Egypt when environmental conditions assist the appearance of the disease (Aly *et al*., 2013). PM requires living tissue for growth and reproduction and thrives in high humidity and moderate temperature environments. PM is one of the major limiting factors of linseed (Dhirhi *et al*., 2017). The importance of this disease may be due to the appearance and the rapid distribution of new races are able to attack the previously resistant cultivar (Aly *et al*., 2017).

Resistance can be induced in plants, which lack the gene for resistance by treatment with some chemicals including phosphate salts (Reuveni *et al*., 2000)

Chemicals such as phosphates, which are used as foliar spray are the target agent for inducing resistance to powdery mildew disease in flax (Reuveni *et al*., 1994).

Disease control in commercial flax is generally depend on the use of fungicides, which negatively affect the environment , the human health, and also the development of pesticide-resistant strains of the powdery mildew pathogen have been reported to decrease the performance of some fungicides. Once resistant strains appear, most of them survive for long periods (Reuveni and Reuveni, 1995) so, there is a need to reduce fungicide use, by alternative methods of disease control. One of such methods is the use of inorganic salts, for instance, phosphates have been shown to induce systemic resistance in many crops (Mitchell and Walters, 2004).

Contrary to the common belief that phosphate salts are effective in inducing local and systemic resistance against powdery mildew. (Mitchell and Walters, 2004) reported that resistance against powdery mildew infection was not effective when the potassium phosphate was applied as a seed treatment or root drench. Descalzo *et al.* (1990) also mentioned that the treatments of dibasic and tribasic

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phosphate salts were ineffective against powdery mildew of cucumber.

In the present study, we evaluated different concentrations of sodium phosphate, potassium phosphate and ammonium phosphate for controlling powdery mildew of flax in an out door pot experiment at Giza Agricultural Research station.

MATERIALS AND METHODS

Plants

Seeds of flax cultivar Giza 12, obtained from Field Crops Research Institute, Agricultural Research Center, Giza, Egypt, were grown on 4 December 2020 outside greenhouse in 25-cm-diameter clay pots containing natural soil and the pots were planted with twenty seeds per pot. This cultivar was found to be susceptible to powdery mildew in previous studies and powdery mildew was allowed to develop naturally.

Foliar application of phosphate salts

An aqueous solutions of the six phosphate salts, Sodium dihydrogen phosphate (NaH_2PO_4) , Sodium monohydrogen phosphate (Na_2HPO_4), Potassium dihydrogen phosphate (KH_2PO_4), Potassium monohydrogen phosphate (K_2HPO_4), Ammonium dihydrogen phosphate ($NH_4H_2PO_4$), Ammonium monohydrogen phosphate ($(NH_4)_2HPO_4$), were prepared (Table 1) and each salt was used in four concentrations (control, 25 mM, 50 mM, 100 mM). Tween 20 was added to each solution as spreader at a rate of 0.5 ml/L. These concentrations were used to determine, which one is the most effective in controlling powdery mildew. In the control, the plants were sprayed only with Tween 20.

The intial application of treatments to plants coincided with the first signs of the disease. Four foliar sprays with phosphate salts were applied to run-off on 18 Febraury, 3 March, 17 March and 2 April 2021 at approximately 14-day intervals. Plants were also fertilized with urea according to recommendations.

Table 1. Phosphate salts used in the present study for controlling powdery mildew of flax.

No.	Name	Chemical formula	Application rate
S1	Sodium dihydrogen phosphate	NaH_2PO_4	25 mM 50 mM 100 mM
S2	Sodium monohydrogen phosphate	Na_2HPO_4	25 mM 50 mM 100 mM
S3	Potassium dihydrogen phosphate	KH_2PO_4	25 mM 50 mM 100 mM
S4	Potassium monohydrogen phosphate	K_2HPO_4	25 mM 50 mM 100 mM
S5	Ammonium dihydrogen phosphate	$NH_4H_2PO_4$	25 mM 50 mM 100 mM
S6	Ammonium monohydrogen phosphate	$(NH_4)_2HPO_4$	25 mM 50 mM 100 mM

Assessment of powdery mildew severity on flax

Disease severity (Aly *et al*., 2013) was measured as percentage of infected leaves/plant in a random sample of 10 plants/pot. At the end of the experiment (21 April 2021), straw and seed weights were recorded for each plant to evaluate the effects of treatments on growth.

Statistical analysis of the data

The experimental design of the pot experiment was a randomized complete block with five replicates. Disease severity ratings of the experiment were transformed into arc sine angles before carrying out the analysis of variance (ANOVA) to produce approximately constant variance. Least significant difference (LSD) was used to compare treatment means. (ANOVA) was carried out by MSTAT-C statistical package.

RESULTS AND DISCUSSION

Analysis of variance (ANOVA) shown in Table 2 indicates that salt was a significant source of variation in disease severity and a very highly significant source of variation in straw weight, while it was a nonsignificant source of variation in seed weight.

Concentration was a very highly significant source of variation in disease severity, seed weight and straw weight.

Salt ×concentration interaction was a highly significant source of variation in disease severity and seed weight while it was a nonsignificant source of variation in straw weight.

Table 2. Analysis of variance of effects of phosphate salts, their concentrations, and the interaction between salt and concentration on powdery mildew severity, seed weight, and straw weight of flax.

Variable and source of variation	D.F	M.S	F. value	P>F
Disease severity				
Replicates	4	273.811	2.134	0.083
Salt (S)	5	380.562	2.966	0.016
Concentration (C)	3	1044.926	8.145	0.000
S×C	15	276.576	2.156	0.013
Error	92	128.292		
Seed weight				
Replicates	4	8386.904	4.474	0.002
Salt (S)	5	3396.908	1.812	0.118
Concentration (C)	3	64955.475	34.650	0.000
S×C	15	4155.068	2.216	0.011
Error	92	1874.635		
Straw weight				
Replicates	4	45398.217	1.881	0.120
Salt (S)	5	118548.033	4.912	0.000
Concentration (C)	3	160584.500	6.653	0.000
S×C	15	24688.740	1.023	0.440
Error	92	24135.878		

Due to the significant of S×C interaction (Table 2) an interaction LSD was used to compare effects of concentrations within salts. These comparison showed that sodium monohydrogen phosphate was the only effective salt in controlling the disease when it was applied at the concentration of 50 mM (Table 3). Other concentrations were either ineffective in controlling the disease or significantly increased disease severity, for example potassium monohydrogen phosphate significantly increased disease severity when it was applied at 100mM

Table 3. Effect of phosphate salts (S), their concentrations (C), and their interaction (S×C) on powdery mildew severity (%) of flax.

	Concentration (C)									
	0		25 mM		50 mM		100 mM		Mean	
Salt	%	Trans.	%	Trans.	%	Trans.	%	Trans.	%	Trans.
S1	80.18	66.52	67.26	55.95	79.18	64.42	89.27	72.88	78.97	64.94
S2	84.00	69.47	72.54	61.60	46.41	42.93	73.00	59.87	68.99	58.47
S3	70.73	57.51	76.00	62.96	81.82	67.73	91.64	73.66	80.05	65.47
S4	70.00	57.27	61.71	51.97	49.25	44.56	92.61	77.85	68.39	57.91
S5	76.00	60.91	83.91	67.58	76.59	61.87	87.25	72.16	80.94	65.63
S6	85.27	68.48	90.91	74.13	75.09	60.87	86.84	71.73	84.53	68.80
Mean	77.70	63.36	75.39	62.37	68.06	57.06	86.77	71.36	76.97	63.54

LSD for S×C interaction (transformed data) = 14.18

Regarding seed yield, concentrations were ineffective in increasing seed yield or significantly reduced it regardless of the tested salt (Table 4).

Table 4. Effect of different concentration of phosphate salts on seed weight of flax (mg/plant).

	Concentration				
Salt	0	25mM	50mM	100mM	Mean
S1	130.40	71.40	32.20	40.00	68.50
S2	160.40	71.60	43.60	45.20	80.20
S3	140.20	84.80	52.80	55.60	83.35
S4	218.60	51.20	54.00	8.80	83.15
S5	145.8	79.40	85.80	110.20	105.30
S6	128.40	119.60	64.80	75.40	97.05
Mean	153.97	79.67	55.53	55.87	86.26

LSD for S×C interaction = 51.75

As to straw weight analysis of variance (Table 2) showed that there was no significant interaction between salt and concentration, so the general means were used to compare between concentrations and salts and it was found that all concentrations significantly decreased straw weight, while salts showed variable effects on straw weight (Table 5).

Table 5. Effect of different concentration of phosphate salts on straw weight of flax (mg/ plant)

	Concentration				
Salt	0	25mM	50mM	100mM	Mean
S1	434.60	474.20	320.00	385.80	403.65
S2	436.40	331.60	312.80	271.40	338.05
S3	487.40	318.00	320.00	427.80	388.30
S4	554.60	240.80	192.00	158.60	286.50
S5	505.40	451.00	466.80	483.00	476.55
S6	588.40	494.80	389.00	462.40	483.65
Mean	501.13	385.07	333.30	364.83	396.08

LSD for concentration = 75.81
LSD for salt = 92.85

The present study clearly demonstrated that compounds such as NaH_2PO_4, KH_2PO_4, K_2HPO_4, $NH_4H_2PO_4$, $(NH_4)_2HPO_4$ were ineffective in controlling powdery mildew on flax in an open-air pot experiment while Na_2HPO_4 (50 mM) was the only effective one as it significantly reduced disease severity by 44.75% ((84-46.4)/84×100).

The significant interaction between salts and concentrations indicated that effects of salts on disease severity and seed weight were affected by the concentration of salts, while in case of straw weight there was nonsignificant interaction and this indicated that the effects of salts on straw weight were not affected by the concentration of salts.

Some workers have reported that the use of phosphate salts as biocompatible fungicides effectively controlled the powdery mildew disease on various crops (Reuveni and Reuveni, 1995), but in the present study, it was found that some treatments increased powdery mildew severity, and some had no effect. Although the mechanism by which these chemicals act is still unknown, the results suggest that the phytotoxic effects of these chemicals may be due to their high concentrations. The results of this study partially agreed with those reported by (Descalzo *et al*., 1990). In their study, the application of K_2HPO_4 was ineffective in controlling powdery mildew disease on cucumber plants. (Reuveni *et al*, 1993).

In conclusion, the present study demonstrated that some phosphate salts are ineffective in controlling the powdery mildew disease of flax when they were applied in high concentrations because they caused phytotoxic effects, which led to increase disease severity and decrease in seed and straw yields.

It seems reasonable to assume that concentration plays a decisive role in determining the efficiency of phosphate salts in controlling flax powdery mildew. Therefore, phosphate salts will be reevaluated in a future study by using lower concentrations. These lower concentrations could be effective in controlling the disease.

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التأثيرات المفيدة و الضارة لإملاح الفوسفات على نباتات الكتان المصابة بالبياض الدقيقى

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استخدمت املاح فوسفات الصوديوم ثنائى الهيدروجين (NaH_2PO_4)، فوسفات الصوديوم احادى الهيدروجين (Na_2HPO_4)، فوسفات البوتاسيوم ثنائى الهيدروجين (KH_2PO_4)، فوسفات البوتاسيوم احادى الهيدروجين (K_2HPO_4)، فوسفات الأمونيوم ثنائى الهيدروجين ($NH_4H_2PO_4$) و فوسفات الأمونيوم احادى الهيدروجين ($(NH_4)_2HPO_4$) لرش أوراق الكتان أربع مرات و ذلك لمقاومة مرض البياض الدقيقى. أظهرت الدراسة أن فوسفات الصوديوم أحادى الهيدروجين (Na_2HPO_4) هو الملح الوحيد الفعال فى مقاومة المرض بكفاءة وصلت الى ٤٤,٧٥٪ عند استخدامة بتركيز ٥٠ mM أما الاملاح الأخرى مثل فوسفات البوتاسيوم أحادى الهيدروجين (K_2HPO_4) فقد احدثت زيادة معنوية فى شدة الاصابة مع نقص فى محصول البذرة و القش. تدل نتائج الدراسة الحالية على ان التركيز يلعب دوراً حاسماً فى تحديد مستوى فاعلية املاح الفوسفات فى مقاومة البياض الدقيقى على الكتان. و على ذلك فإن إعادة تقييم هذة الاملاح مستقبلاً باستعمال تركيزات أقل قد يؤدى إلى فاعلية فى مقاومة المرض.