

THE EFFECTS OF ORGANIC ACIDS TREATMENT ON GERMINATION, VIGOUR AND HEALTH OF *ZINNIA* (*Zinnia elegans* Jacq.) SEEDS

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Abstract. Organic acids are known for their antibacterial and antifungal properties. The purpose of the research was to study the effect of acetic, ascorbic, citric and lactic acid treatments on zinnia seed germination, vigour and infestation with fungi. Two seed samples, cultivars 'Jowita' and 'Scarlet Flame', varying in fungi occurrence intensity, were treated with organic acids solutions at concentrations of 1.0, 2.5 and 5.0%. Controls were untreated seeds, seeds treated with fungicide Penncozeb 80 WP, and seeds soaked for 30 min in distilled water. Acetic acid in the highest extent limited fungal occurrence on seeds, however negatively affected seed germination and vigour. Ascorbic and citric acids had no effect on the total seed infestation with fungi. Moreover, the acids significantly stimulated growth of *Botrytis cinerea* on the seeds. Lactic acid decreased the number of seeds infected with fungi, especially with *Alternaria zinniae* and *Fusarium* spp., however at the highest concentration negatively affected germination parameters and deteriorated seed vigour.

Key words: acetic acid, ascorbic acid, citric acid, lactic acid, zinnia seeds quality, seed-borne fungi

INTRODUCTION

Zinnia elegans Jacq. is a worldwide popular ornamental plant growing for cut flowers and flowerbeds. Łacicowa et al. [1979] reported that *Alternaria zinniae* M.B. Ellis, *Fusarium culmorum* (W.G. Smith) Sacc., *F. solani* (Mart.) Sacc., *F. oxysporum* Schlecht. and *Sclerotinia sclerotiorum* (Lib.) de Bary had been in the most cases responsible for severe damages of zinnia plants in the field. Richardson [1990] listed the following fungi as seed transmitted pathogens of zinnia: *A. zinniae*, *Botrytis cinerea* Pers. ex Fr., *Colletotrichum acutatum* Simmonds, *Erisiphe cichoracearum* DC., *Glomerella cingulata* (Stoneman) Splaud & H. Schrenk, *Phyllosticta* sp. and *Rhizocto-*

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nia solani Kühn. *Alternaria* blight caused by *A. zinniae* is a common seedborne disease of garden zinnia, expressed in spotting of petals, foliage and stems, and root rot [Dimock and Osborne 1943, Palacios et al. 1991, Wu and Yang 1992]. Nearly all lots of zinnia seeds may be infected with this pathogen in some years. *Alternaria alternata*, *B. cinerea*, *Fusarium* spp. and *Penicillium* spp. were also frequently detected in zinnia seeds produced in Poland [Łacicowa et al. 1991, Szopińska and Tytkowska 2009].

In ecological farming, natural antimicrobial compounds, such as organic acids, can be used for seed disinfection as an alternative, or in combination with physical treatment. Functions of ascorbic acid in plants are largely related to redox properties associated with this molecule. The acid is rapidly synthesized during seed germination and continues to be produced in regions of active growth throughout the life of the plant, but seldom is accumulated beyond 100 mg% of fresh weight [Loewus 1999]. The antibacterial activity of ascorbic acid had been reported from the beginning of the twentieth century. The scientists found that inhibition of *Bacillus subtilis*, *Escherichia coli* and *Proteus vulgaris* by ascorbic acid was counteracted by the presence of reducing agents and by substances which catalyzed the breakdown of hydrogen peroxide. They concluded that the inhibition was due to hydrogen peroxide formed during auto-oxidation of ascorbic acid [Myrvik and Volk 1954]. Citric acid as well as ascorbic acid can neutralize harmful oxygen radicals. Aml El-Saidy and Abd El-Hai [2011] found that the acids effectively controlled fungi, improved seed germination and reduced deterioration of peanut seeds under storage.

Acetic acid is a metabolic intermediate that occurs naturally in many fruits. Vapour of this acid controlled common bunt in wheat, reducing significantly level of plant infection in the field [Sholberg et al. 2006]. Fumigation of high moisture seeds of canola, corn, rice, and wheat with acetic acid effectively prevented growth of *Aspergillus flavus* Link ex Fries during storage [Sholberg and Gaunce 1996]. Acetic acid was also successfully used to eliminate *Escherichia coli* [Lang et al. 2000] and *Salmonella enterica* [Pao et al. 2008] from alfalfa sprouts.

Different strains of *Lactobacillus* produced a broad-spectrum of antifungal and antibacterial compounds, e.g. the pH reducing fermentation products lactic and acetic acid, as well as hydrogen peroxide, formic acid, propionic acid, and diacetyl [Schnürer and Magnusson 2005]. These products, especially lactic acid, the major *Lactobacillus* metabolite, have been commonly used as food preservatives [De Muynck et al. 2004, Latila et al. 2002, Lavermicocca et al. 2003, Magnusson and Schnürer 2001, Magnusson et al. 2003, Rouse et al. 2008, Sathe et al. 2007, Schnürer and Magnusson 2005, Ström et al. 2005].

The aim of the experiment was to investigate effect of four organic acids i.e. acetic, ascorbic, citric, and lactic acid, on germination, vigour and health of zinnia seeds.

MATERIALS AND METHODS

Two zinnia seed samples, cultivars 'Jovita' (sample I) and 'Scarlet Flame' (sample II) obtained from seed company TORSEED in Toruń were used in the study. Seeds were treated with acetic, ascorbic, citric and lactic acids, produced by Sigma Chemical

Co. Fungicide Penncozeb 80 WP (a.i. mancozeb 80%) was used as an alternative chemical control.

Germination, vigour and seed health tests were performed for: untreated seeds (control I), seeds treated with fungicide 5 g kg⁻¹ of seeds (control II), seeds soaked for 30 min in distilled water (control III), and seeds soaked for 30 min in 1.0, 2.5 and 5.0% solutions of acetic, ascorbic, citric and lactic acid, respectively.

Seed germination test. For germination test percentage of normal seedlings (germination capacity) and abnormal seedlings, both deformed and diseased, were determined after four and ten days according to the ISTA rules [International Rules for Seed Testing 2006]. Twelve replicates of 25 seeds from each treatment (300 seeds) were placed in Petri dishes containing six layers of moistened blotters and incubated at 20°C, in darkness. Additionally, the total number of germinating seeds (G_{max}) was determined on the base of number of seeds with visible radicle counted daily.

Seed vigour test. Twelve replicates of 25 seeds from each treatment (300 seeds) were incubated under the same conditions as described in the previous test to evaluate seed vigour. Radicle protrusion was scored daily for ten days. The germination rates, i.e.: T_1 – time to 1% of G_{max} , MGT – mean germination time and U_{75-25} – time between 25 and 75% of G_{max} , were evaluated.

Mycological analysis. The seed health analysis was performed on 200 seeds from each treatment. The seeds were plated on six layers of moistened blotter placed in Petri dishes at the rate of 10 seeds per dish, and incubated in darkness at 20°C for one day, at -20°C for 20 h, and then for eight days at 20°C, under 12 h alternating cycles of NUV light and darkness. After incubation the fungi were identified on the basis of their growth and sporulation using a stereomicroscope and, if necessary, compound microscope [Machado et al. 2002, Mathur and Kongsdal 2003]. Additionally the number of seeds free from fungi was evaluated.

Statistical analysis. SeedCalculator version 2.1 software [Jalink and Van der Schoor 1999] was applied to analyze total germination and vigour data. All results were compared by means of variance analysis followed by the Duncan's test.

RESULTS

Seed germination. Treatment of zinnia seed sample I with acetic acid at concentration 2.5 and 5.0% significantly decreased total number of germinating seeds (G_{max}). Acetic acid in all concentrations, as well as 5.0% citric acid, and 2.5 and 5.0% lactic acid negatively influenced this parameter in sample II. Germination capacity of seeds of sample I decreased significantly after treatment with 2.5 and 5.0% acetic acid and 5.0% lactic acid. These acids, regardless of concentration, as well as 5.0% ascorbic acid and citric acid at 2.5 and 5.0% concentrations negatively effected germination capacity of seeds of sample II. Improvement of the parameter was observed only in sample I after fungicide treatment. In this sample the increase of the number of abnormal deformed seedlings and the decrease of the number of diseased seedlings were observed after Penncozeb 80 WP application. Treatment of both sample seeds with acetic acid in 2.5% and 5.0% concentrations negatively affected the number of deformed seedlings. More-

over, in sample I the highest concentration of lactic acid, and in sample II 1.0% acetic acid and 5.0% citric acid increased number of these seedlings. Considerably lower number of abnormal diseased seedlings was noted in both samples only after 5.0% acetic acid treatment. The opposite effect was observed, when seeds of sample I were treated with 1.0 and 5.0% solution of ascorbic acid, and seeds of sample II were soaked in 5.0% solution of ascorbic acid and 2.5 and 5.0% solutions of citric and lactic acids (tab. 1).

Seed vigour. Treatment of seed sample I with fungicide, 2.5% ascorbic acid, and acetic acid regardless of concentration, prolonged the time needed for 1% germination of the total number of germinating seeds, expressed as a T_1 parameter. Deterioration of this parameter was observed in sample II only after acetic acid treatment, regardless of concentration.

Acetic acid at all concentrations, as well as fungicide and 5.0% lactic acid, affected negatively mean germination time of both samples. Moreover, 1.0, 2.5 and 5.0% ascorbic acid deteriorated the parameter in sample II. Treatment of zinnia seeds sample I with acetic acid at concentrations 2.5 and 5.0% negatively affected uniformity of germination (U_{75-25}). However, deterioration of this parameter was observed in sample II after the treatment with 1.0, 2.5 and 5.0% acetic acid and 5.0% lactic acid (tab. 2).

Mycological analysis. Ten fungal species and unidentified one belonging to seven genera were associated with the seeds. Among them: *Alternaria alternata* (Fr.) Keissler, *A. zinniae* M.B. Ellis, *Botrytis cinerea* Pers. ex Pers., *Gonatobotrys simplex* Corda and species of *Cladosporium* and *Fusarium* occurred most frequently (tab. 3 and 4). The other fungi: *Acremoniella atra* (Corda) Sacc., *Acremonium strictum* W. Gams, *Aspergillus niger* van Tieghem, *Epicoccum purpurascens* Ehrenb. ex Schlecht., *Rhizopus nigricans* Ehrenberg, *Trichothecium roseum* (Pers.) Link ex S.F. Gray, and unidentified species of *Colletotrichum*, *Mucor*, *Papulaspora*, *Penicillium*, and *Ulocladium* were noted occasionally, many a time occurring on the seeds of only one sample. Moreover, infestation of the seeds with these saprotrophic fungi did not exceed 0.5–1.5%, regardless of the treatment (data not shown).

The samples differed significantly in terms of the number of fungi species and percentage of infested seeds. Sample I was characterized by very high percent (57.0) of infection with *A. zinniae*, however, the pathogen infected only 0.5% of seeds in sample II. On the other hand sample II showed higher seed infection with *B. cinerea* and *Fusarium* spp. The fungi were observed in 0.5% and 73.0% of sample I seeds, and 5.5% and 97.5% of sample II seeds, respectively.

Acetic acid, at all concentrations, and lactic acid at higher concentrations comparably to fungicide controlled growth of *A. zinniae* in sample I, and *A. alternata* in both samples. Moreover, infection with *A. zinniae* decreased significantly if seeds of sample I were soaked in 1.0% solution of lactic acid and in 2.5 and 5.0% solutions of citric acid. Stimulation of the growth of *A. alternata* on the seeds of sample II was observed after treatment with 1.0 and 2.5% ascorbic acid and citric acid, regardless of concentration. Moreover, an increase of the number of seeds infected with *A. zinniae* was noted in sample I, if the seeds were treated with 1.0% ascorbic acid. Seed treatment with ascorbic and citric acids increased significantly the number of seeds infected with *B. cinerea* in both samples – the higher concentration the higher infection level. This effect was

Table 1. Effects of organic acids on zinnia seed germination

Treatment	Percent of germinating seeds			Germination capacity, %			Abnormal deformed seedlings, %			Abnormal diseased seedlings, %		
	sample I	sample II	sample I	sample I	sample II	sample I	sample I	sample II	sample I	sample II	sample I	sample II
Control I ¹	84.0 cd	91.0 f	47.0 def	68.7 g	0.3 ab	1.7 abc	30.3 bcd	19.3 bcd				
Control II ²	86.7 d	89.7 f	68.3 g	69.0 g	3.3 c	0.7 a	11.7 a	16.7 bcd				
Control III ³	86.0 d	88.3 ef	54.0 def	66.3 fg	0.3 ab	2.0 abc	25.3 b	18.7 bcd				
Acetic acid 1% ⁴	84.0 cd	78.3 cd	53.0 def	32.7 c	0.7 abc	13.0 ef	23.0 b	21.0 bcd				
Acetic acid 2.5% ⁴	63.7 b	36.0 b	28.3 b	4.0 b	8.3 d	19.3 f	28.3 bc	13.3 ab				
Acetic acid 5% ⁴	26.3 a	14.0 a	4.7 a	0 a	10.3 de	11.7 e	12.7 a	8.3 a				
Ascorbic acid 1% ⁵	85.3 d	88.3 ef	40.0 bcd	68.3 fg	2.0 bc	1.0 ab	44.0 e	16.0 bc				
Ascorbic acid 2.5% ⁵	86.7 d	90.3 f	42.7 cde	67.0 fg	1.0 abc	0.7 ab	44.0 de	19.7 bcd				
Ascorbic acid 5% ⁵	82.0 cd	87.0 ef	40.7 bcd	49.7 de	1.3 abc	2.0 abc	44.7 e	33.3 ef				
Citric acid 1% ⁶	88.0 d	86.7 ef	58.0 fg	61.0 efg	0.7 abc	1.3 ab	25.7 b	23.7 cde				
Citric acid 2.5% ⁶	87.0 d	87.7 ef	40.3 bcd	49.7 de	2.0 bc	2.7 abcd	43.0 de	32.7 ef				
Citric acid 5% ⁶	83.3 cd	83.0 de	42.3 cde	36.3 c	0 a	6.3 de	40.0 cde	40.7 f				
Lactic acid 1% ⁷	85.7 d	86.0 ef	55.7 ef	57.3 ef	0 a	1.7 abc	23.7 b	26.3 de				
Lactic acid 2.5% ⁷	85.7 d	82.0 de	48.3 def	39.3 cd	2.0 abc	3.3 bcd	25.3 b	31.3 ef				
Lactic acid 5% ⁷	75.3 c	73.3 c	31.7 bc	32.3 c	16.0 e	5.3 cd	29.0 bc	33.3 ef				

¹ Control I – untreated seeds

² Control II – seeds treated with fungicide Penncozeb 80 WP – 5 kg⁻¹ of seeds

³ Control III – seeds soaked for 30 min in distilled water

⁴ Seeds soaked for 30 min in 1.0, 2.5 and 5.0% solutions of acetic acid

⁵ Seeds soaked for 30 min in 1.0, 2.5 and 5.0% solutions of ascorbic acid

⁶ Seeds soaked for 30 min in 1.0, 2.5 and 5.0% solutions of citric acid

⁷ Seeds soaked for 30 min in 1.0, 2.5 and 5.0% solutions of lactic acid

Means in columns followed by the same letter do not differ significantly at $\alpha = 0.05$ according to Duncan's test

Table 2. Effects of organic acids on zinnia seed vigour (days)

Treatment	T ₁ ¹		MGT ²		U _{75,25} ³	
	sample I	sample II	sample I	sample II	sample I	sample II
Control I	0.00 ab	0.11 a	1.06 ab	0.77 ab	1.19 a	0.55 a
Control II	0.82 de	0.27 a	1.74 e	1.18 cd	0.72 a	0.73 a
Control III	0.00 a	0.03 a	0.86 a	0.65 a	1.15 a	0.62 a
Acetic acid 1%	0.69 cde	0.57 b	2.11 f	2.90 e	1.18 a	1.85 b
Acetic acid 2.5%	0.41 cde	0.98 b	3.45 g	4.44 f	2.56 b	2.78 c
Acetic acid 5%	0.88 e	0.94 c	4.80 h	4.72 g	3.17 c	2.98 c
Ascorbic acid 1%	0.50 bcd	0.00 a	1.62 de	0.87 abc	0.89 a	1.12 a
Ascorbic acid 2.5%	0.85 cde	0.09 a	1.52 cde	0.91 abc	0.53 a	0.72 a
Ascorbic acid 5%	0.68 bcde	0.18 a	1.72 e	1.05 bc	0.81 a	0.71 a
Citric acid 1%	0.10 ab	0.04 a	1.20 ab	0.78 ab	1.01 a	0.71 a
Citric acid 2.5%	0.64 abcd	0.04 a	1.30 bc	0.74 ab	0.52 a	0.67 a
Citric acid 5%	0.62 abcd	0.04 a	1.39 bcd	0.85 ab	0.64 a	0.80 a
Lactic acid 1%	0.26 abc	0.00 a	1.33 bcd	0.67 ab	0.89 a	0.94 a
Lactic acid 2.5%	0.65 bcde	0.05 a	1.32 bcd	0.99 bc	0.53 a	0.89 a
Lactic acid 5%	0.41 abc	0.06 a	1.71 e	1.43 d	1.14 a	1.34 b

¹ Time to 1% of total number of germinating seeds

² Mean germination time

³ Time between 25 and 75% of total number of germinating seeds

For further explanations see table 1

Table 3. Effects of organic acids on the zinnia seeds infestation with fungi – sample I

Treatment	Percent of infested seeds							Seeds free from fungi, %
	<i>Alternaria alternata</i>	<i>Alternaria zinniae</i>	<i>Botrytis cinerea</i>	<i>Cladosporium</i> spp.	<i>Fusarium</i> spp.	<i>Gonotobotrys simplex</i>		
Control I	87.0 cd	57.0 d	0.5 a	52.0 cd	73.0 fg	71.0 f	0 a	
Control II	40.5 a	24.0 ab	1.0 a	3.0 a	30.5 c	0 a	21.5 de	
Control III	86.5 cd	59.0 de	1.0 a	53.0 cd	74.5 fg	70.5 f	0.5 a	
Acetic acid 1%	45.0 a	38.5 c	0.5 a	5.0 a	29.0 c	0 a	19.0 cd	
Acetic acid 2.5%	47.5 ab	30.5 abc	0.5 a	4.5 a	17.0 b	0 a	19.0 cd	
Acetic acid 5%	39.0 a	27.0 ab	0 a	2.5 a	10.0 ab	0 a	30.5 f	
Ascorbic acid 1%	89.0 cd	70.0 e	6.0 b	57.0 cde	80.0 g	72.5 f	0 a	
Ascorbic acid 2.5%	90.0 cd	51.5 d	18.5 de	69.0 e	64.5 ef	63.5 f	0 a	
Ascorbic acid 5%	92.0 d	52.5 d	27.5 ef	62.5 de	58.0 de	52.0 e	0 a	
Citric acid 1%	88.5 cd	63.0 de	12.5 cd	61.5 de	74.5 fg	64.5 f	0 a	
Citric acid 2.5%	92.5 d	39.5 c	32.0 fg	69.0 e	49.5 d	40.0 d	0 a	
Citric acid 5%	92.5 d	32.5 bc	40.0 g	66.5 de	53.0 de	14.0 c	0 a	
Lactic acid 1%	80.5 c	33.0 bc	13.0 cd	43.5 c	27.0 c	15.5 c	3.0 b	
Lactic acid 2.5%	58.5 b	23.0 ab	7.0 bc	20.0 b	8.0 a	4.5 b	13.0 c	
Lactic acid 5%	41.0 a	21.5 a	3.5 ab	15.0 b	6.0 a	2.0 b	27.5 ef	

For explanation see table 1

Table 4. Effects of organic acids on the zinnia seeds infestation with fungi – sample II

Treatment	Percent of infested seeds						Seeds free from fungi, %
	<i>Alternaria alternata</i>	<i>Alternaria zinniae</i>	<i>Botrytis cinerea</i>	<i>Cladosporium</i> spp.	<i>Fusarium</i> spp.	<i>Gonotobotrys simplex</i>	
Control I	79.0 fg	0.5 ab	5.5 ab	39.5 c	97.5 f	23.5 e	0 a
Control II	40.5 abc	0 a	10.5 bcd	11.0 b	60.0 c	0.5 ab	13.5 b
Control III	78.0 fg	0.5 ab	6.5 abc	32.5 c	95.5 ef	18.0 de	0 a
Acetic acid 1%	60.5 de	0.5 ab	8.0 abc	2.0 a	66.5 c	0 a	0.5 a
Acetic acid 2.5%	53.0 cd	0.5 ab	8.5 bc	2.5 a	57.5 c	0 a	9.5 b
Acetic acid 5%	38.5 ab	0 a	5.5 ab	2.0 a	22.5 a	0 a	33.0 c
Ascorbic acid 1%	95.0 h	1.0 b	14.5 cde	61.0 d	91.0 de	21.0 de	0 a
Ascorbic acid 2.5%	90.5 h	0 a	24.0 efg	58.5 d	91.0 de	16.5 de	0 a
Ascorbic acid 5%	87.0 gh	0 a	26.5 fgh	66.0 de	88.5 d	15.0 cd	0 a
Citric acid 1%	92.0 h	0 a	23.0 efg	71.0 def	90.5 de	11.0 c	0 a
Citric acid 2.5%	93.5 h	0 a	35.5 gh	80.0 f	83.5 d	0 a	0 a
Citric acid 5%	90.0 h	0 a	38.5 h	76.0 ef	91.5 def	2.0 b	0 a
Lactic acid 1%	67.5 ef	0.5 ab	19.0 def	65.0 de	36.0 b	1.5 ab	0.5 a
Lactic acid 2.5%	46.0 bc	0 a	8.5 abc	32.0 c	23.5 a	0 a	16.0 b
Lactic acid 5%	28.5 a	0 a	2.5 a	14.5 b	13.0 a	0 a	37.5 c

For explanation see table 1

also observed if the seeds were treated with lactic acid, especially at the lowest concentration. Fungicide and acetic acid treatment strongly decreased seed infestation with *Cladosporium* spp. Similar effect was observed if seeds of sample I were treated with 2.5 and 5.0% lactic acid, and seeds of sample II were soaked in 5.0% solution of this acid. Infestation with *Cladosporium* spp. increased significantly if seeds were treated with 2.5% ascorbic and citric acid in sample I, whereas both acids at 1.0, 2.5 and 5.0% concentration, and 1.0% lactic acid stimulated growth of these fungi in sample II. Acetic and lactic acids at higher concentrations as well as Penncozeb 80 WP, or even better than the fungicide, controlled *Fusarium* spp. on zinnia seeds. The growth of the fungi was also limited by 5.0% ascorbic acid, and 2.5 and 5.0% citric acid in sample I, and by 1.0 and 2.5% citric acid and ascorbic acid at all concentrations in sample II. All acids as well as fungicide limited percentage of seeds infested with *Gonatobotrys simplex* in both samples. However, ascorbic acid was effective only at the highest concentration in both samples, whereas citric acid at the lowest concentration did not affected growth of this fungus in sample I.

Fungicide treatment and soaking seeds in acetic and lactic acid regardless of concentration in sample I, as well as treatment of seeds with 1.0 and 2.5% solutions of these acids in sample II increased the number of seeds free from fungi.

DISCUSSION

Organic acids are known for years for their antibacterial and antifungal properties which have been widely used in foodstuff industry and agriculture [De Muynck et al. 2004, Laitila et al. 2002, Lavermicocca et al. 2003, Pao et al. 2008, Sathe et al. 2007, Scholberg and Gaunce 1995, 1996, Tripathi and Dubey 2004].

Acetic acid applied as vapour to high moisture content canola, corn, rice and wheat inoculated with conidia of *Aspergillus flavus* effectively prevented growth of the fungus. Wheat seeds fumigated with acetic acid at 0.78 mL kg⁻¹ of seed characterized maximum germination and no infection after storage for 102 days at 20°C [Sholberg and Gaunce 1996]. Sholberg et al. [2006] inoculated grains of highly susceptible wheat cultivar with bunt spores (*Tilletia tritici* and *T. laevis*) and then fumigated with 2 and 4 g kg⁻¹ acetic acid vapour. Fumigation reduced field infection levels of common bunt as effectively as standard seed-treatment fungicide. However, some reduction in tiller numbers was associated with the acetic acid treatments especially at the 4 g kg⁻¹ rate. Sholberg and Gaunce [1995] found that low concentrations of acetic acid vapour were extremely effective for control of *B. cinerea* conidia on apple fruit without phytotoxic effect. In present experiment acetic acid significantly reduced seed infestation with *Alternaria* spp., *Cladosporium* spp., *Fusarium* spp. and *G. simplex*, however, did not control seed infection with *B. cinerea*. Moreover, the acid decreased total number of germinating seeds and germination capacity, increased number of abnormal deformed seedlings, and prolonged germination at all tested concentrations. Pasini et al. [1997] studied effectiveness of acetic acid against rose powdery mildew (*Sphaerotheca pannosa* var. *rosae*) in glasshouses. The authors reported that white vinegar, applied at 5.0 and 10.0% gave good disease control, but acetic acid at 0.25 and 0.5% was phytotoxic.

Acetic and lactic acid combined with hypochlorite treatment were used by Lang et al. [2000] for eliminating *Escherichia coli* O157:H7 from alfalfa seeds prior sprouting. The authors observed that germination of seeds was not adversely affected by any of the treatments (> 90%), regardless of quite high concentration of the acids (5.0%). Pao et al. [2008] achieved elimination of *Salmonella enterica* in alfalfa and mung bean sprouts after 4 and 16 hours immersing in 5% acetic acid. However, similar treatments with citric acid were ineffective.

Lactic acid bacteria have been exploited for decades for their antibacterial activity. More recently, they have also got special attention because of their antifungal potential [Rouse et al. 2008]. Magnusson and Schnürer [2001] observed strong inhibitory activity of *Lactobacillus coryniformis* subsp. *coryniformis* strain Si3 in dual-culture agar plate assays against *Aspergillus fumigatus*, *A. nidulans*, *Penicillium roqueforti*, *Mucor hiemalis*, *Talaromyces flavus*, *Fusarium poae*, *F. graminearum*, *F. culmorum*, and *F. sporotrichioides*. In the other experiment the growth of the indigenous *Fusarium* spp. was restricted by the addition of *Lactobacillus plantarum* E76 to the steeping water in the barley malting process. However, the antifungal effect was greatly dependent on the contamination level and the fungal species/strains presented on barley in different years [Laitila et al. 2002]. Ström et al. [2005] studied growth of *Aspergillus nidulans* in the presence of antifungal substances produced by *Lactobacillus plantarum* MiLAB: 100 mM lactic acid, 20 mM 3-phenyl lactic acid and 10 mg ml⁻¹ cyclo(L-Phe-L-Pro). The authors observed that all three compounds affected growth of the fungus at the concentrations used. Magnusson et al. [2003] reported that the degree of fungal inhibition by lactic acid bacteria was not only related to production of lactic and acetic acid, but several other active compounds, suggesting a highly complex nature of antifungal activity of *Lactobacillus coryniformis* and *L. plantarum*. In present experiment the lactic acid especially at 2.5 and 5.0% concentration strongly inhibited growth of *Alternaria alternata*, *A. zinniae*, *Fusarium* spp. and *Gonatotryps simplex* on zinnia seeds. However, the lowest concentration of the acid stimulated growth of *Botrytis cinerea* and *Cladosporium* spp. Moreover, 5.0% solution of lactic acid negatively affected germination and vigour of the seeds, especially in sample II.

Van der Wolf et al. [2008] examined effect of treatment with different concentrations of organic acids (from 0.5 to 10.0%) on cabbage seed-associated bacteria. Disinfection with lactic acid at 0.5% resulted in the reduction of the bacterial count by more than 99%. However, acetic and ascorbic acids reduced the bacterial count by more than 99% at concentrations of 2.5% and higher, but not at 0.5%. *In vitro* assays showed slight activity of acetic and lactic acid against *Alternaria dauci*, whereas ascorbic and citric acids even at the highest 10.0% concentration showed no activity against *A. dauci* and *Botrytis aclada* *in vitro*. In the present experiment ascorbic acid, as well as citric acid treatment, slightly reduced seed infection with *Fusarium* spp., but significantly stimulated growth of *Botrytis cinerea* on zinnia seeds. The increase of the number of seeds on which *A. alternata* and *Cladosporium* spp. occurred was also observed in one of tested samples, while Aml El-Saidy and Abd El-Hai [2011], which studied effect of citric acid on infestation of peanut seeds with fungi under storage, recorded an inhibitory effect of this acid on the presence of species representing nine genera i.e., *Alternaria*, *Aspergillus*, *Botrytis*, *Cephalosporium*, *Fusarium*, *Penicillium*, *Rhizoctonia*,

Rhizopus and *Verticillium* in all storage periods (2, 4 and 6 months). However, there was no effect of citric acid on total fungal population counts at 0 time of storage. Meanwhile, the interactions between storage periods and any of the treatments had no significant effect on germination criteria. The authors also found that ascorbic and citric acid, especially at lower concentrations, did not affect most of the germination and vigour parameters.

The strong antifungal activity of acetic and lactic acid seems to be very promising for further studies on zinnia seed treatment. The new method of acids application, proper doses and time of seed exposition should be found to maintain benefits of the treatment, without affecting germination and vigour parameters.

CONCLUSIONS

1. Acetic acid in the highest extent reduced seed infestation with fungi, however negatively affected seed germination and vigour.

2. Generally, ascorbic and citric acid had no influence on germination and vigour parameters, however stimulated growth of some fungi, especially *B. cinerea*.

3. Lactic acid, especially at the higher doses, decreased the number of seeds infected with fungi, especially *A. zinniae* and *Fusarium* spp. The acid at 5.0% concentration decreased seed germination capacity and prolonged mean germination time.

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WPLYW TRAKTOWANIA NASION CYNII (*Zinnia elegans* Jacq.) KWASAMI ORGANICZNYMI NA ICH KIELKOWANIE, WIGOR I ZDROWOTNOŚĆ

Streszczenie. Kwasy organiczne są znane ze swych antybakteryjnych i antygrzybowych właściwości. Celem doświadczenia było określenie wpływu traktowania nasion cynii kwasami: octowym, askorbinowym, cytrynowym i mlekowym na ich kielkowanie, wigor i zasiedlenie przez grzyby. Dwie próby nasion cynii odmian 'Jowita' i 'Scarlet Flame', różniące się nasileniem występujących na nich grzybów, traktowano roztworami kwasów organicznych o stężeniu 1,0, 2,5 i 5,0%. Kombinację kontrolną stanowiły nasiona nietraktowane, nasiona traktowane fungicydem Penncozeb 80 WP i nasiona moczone w wodzie destylowanej przez 30 min. Kwas octowy w największym stopniu ograniczał występowanie grzybów na nasionach, jakkolwiek negatywnie wpływał na kielkowanie i wigor nasion. Kwasy askorbinowy i cytrynowy nie miały wpływu na ogólne zasiedlenie nasion przez grzyby. Ponadto, kwasy te istotnie stymulowały wzrost *Botrytis cinerea* na nasionach. Kwas mlekowy zmniejszał liczbę nasion porażonych przez grzyby, zwłaszcza przez *Alternaria zinniae* i *Fusarium* spp., niemniej w najwyższym stężeniu negatywnie wpływał na parametry kielkowania i pogarszał wigor nasion.

Słowa kluczowe: kwas octowy, kwas askorbinowy, kwas cytrynowy, kwas mlekowy, jakość nasion cynii, grzyby przenoszone z nasionami

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