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Research Article
Effect of some Evaporation Matters on Storability of Sunflower (*Helianthus annuus* L.) Seed

[1]Aml E.A. El-Saidy and [2]K.M. Abd El-Hai

[1]Department of Seed Technology Research, Field Crop Research Institute, Agricultural Research Center, Giza, Egypt
[2]Department of Leguminous and Forage Crop Diseases, Plant Pathology Research Institute, Agricultural Research Center, Giza, Egypt

Abstract

Background and Objective: This study focuses on finding compounds that are safe to humans and environment, such as propionic and acetic acids that may provide an alternative control of seed-borne pathogens and decrease seed deterioration during storage. The objectives of this study were to reduce sunflower seed deterioration and improve the viability of sunflower seed using environmentally safe organic acids. **Materials and Methods:** Propionic and acetic acids were applied on sunflower seed at different concentrations under laboratory conditions during different storage periods. After 6 months storage period, the viability of sunflower seed as well as morphological and physiological characteristics of seedlings were evaluated under greenhouse conditions. Laboratory experiment was conducted in a factorial completely randomized design and randomized complete block design for greenhouse experiment. **Results:** Propionic and acetic acids at different concentrations showed inhibitory effects on the presence of different fungal genera in all storage periods. Propionic acid was most effective followed by acetic acid. Increasing storage periods from 0-6 months significantly decreased germination percentage, germination energy, seedling characters, survived healthy seedlings and seed oil and protein percentages but dead and rotted seeds, as well as rotted seedlings were increased. Treating sunflower seeds with propionic acid (100%) improved germination criteria, seedling characters and seed chemical characters as well as survival seedlings and minimized the dead seeds, rotted seeds and rotted seedlings as compared with the control under all storage periods. Under greenhouse conditions, the maximum growth parameter and physiological characters (chlorophylls a, b, carotenoids and total phenols) were recorded from seed treated with 100% propionic acid after 6 months of storage. **Conclusion:** It may be concluded that propionic and acetic acids vapors can have considerable fungicidal activity against sunflower pathogens and improve seed viability. Therefore, it is recommended using 100% propionic acid to reduce deterioration and seed-borne pathogens of sunflower under storage conditions.

Key words: Sunflower, storage, propionic acid, acetic acid, germination, seedling vigor, viability

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Corresponding Author: Aml E.A. El-Saidy, Department of Seed Technology Research, Field Crop Research Institute, Agricultural Research Center, Giza, Egypt Tel: 00201001064654

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Sunflower (*Helianthus annuus* L.) is one of the most widely cultivated oil crops in the world. It's belonging to family composite, it's a short duration crop and can be grown twice a year[1]. Sunflower seed oil content ranges from 40-50% which contains a high linoleic acid, an unsaturated fatty acid that aids in lowering the cholesterol level in the blood. It also contains fat-soluble vitamins A, D, E, K and good for heart patients. Generally, this oil is used for human food, livestock feed and other industrial products such as soft margarines, salad oil, paint and emulsion industries[2-4]. Seed deterioration during storage is a natural phenomenon and the seeds tend to lose viability even under ideal storage conditions. The chemical composition of oil crops such as sunflower are related to specific processes occurring in seed during storage[5].

Oily seeds have limited longevity due to its specific chemical composition, declining trend of total oil content and seed germination can be observed during storage. Oxidation of fatty acids during storage period is the main reason for rapid deterioration of oil seeds[6,7]. Seed oil content in sunflower decreased by 8.53% and seed vigor decreased by 57.1% affected by storage longevity for storage from 2002-2006[8]. They concluded that, if suitable storage conditions are not provided, quality and quantity losses increase. Moreover, storage conditions and the period of storage have large influence on the quality of sunflower seed. Period of storage had a negative effect on seed germination[9]. They stated that sunflower seed germination declined significantly after a year of storage period. The storage period of sunflower seed can be decreased by longer storage time, hence, reduction in oil content while, free fatty acids content of crude oil increased in longer storage time[7]. Fatty acids type presented in an oil and in particular their number of double bonds, determine the type and extent of chemical reactions which occur during storage time[6]. Unsaturated fatty acids are very important for the stability of oils because of the chemical reactions occurring at the double bonds. The deterioration of sunflower seeds under storage period for 4 months can be observed by the reduced oil content, the disintegration of cellular components and from the analysis of the dehydrogenase, superoxide dismutase, catalase and malate dehydrogenase enzymes[10]. They added that *Aspergillus* sp. and *Penicillium* sp. occurred regardless of storage conditions. So, sunflower seed storage demands special care due to high oil content which can easily processes that can lead to loss of germination and viability.

The deterioration of seed during storage period is a major problem due to the damage of seed viability and vigor in the presence of microorganisms within high relative humidity[11]. In addition, many factors limit sunflower planting, growth and productivity include the partial interruption in harvested heads number due to broken plants caused by fungal infection[12]. Sunflower attacks by more than 35 pathogens, fungal diseases causes the majority of losses. Fungal infection during harvest period of flower heads leads to more rapid seed deterioration during storage. It increase loss of seed vigor, germination and viability[12,9]. Application of seed treatment is the most widely in the traditional one of protecting the germinating seedling against seed and soil-borne fungi in the period immediately after planting. Also, the uses and expectation of seed treatments are greater today due to the impact of environmental regulations that have either banned or restricted the use of the potential to control different pathogens and provide plant protection well into the growing season[13].

The present study focuses on finding compounds that are safe to humans and the environment, e.g., propionic and acetic acids and may provide an alternative control of many soils and seed-borne pathogens. Short-chain volatile fatty acids such as acetic, propionic, butyric, valeric and caproic acids are metabolic products of bacterial anaerobic fermentation in liquid manures[14]. These compounds kill human and animal pathogens[15] and food spoilage organisms[16]. Acetic and propionic acids are universal metabolic intermediaries and occur in plants and animals[17]. Therefore, they were commonly used in food manufacturers as an antimicrobial preservative or acidulates in a variety of industrial food products[18]. Acetic acid inhibits many species of bacteria, yeasts and molds. It is known to be an effective antifungal agent, particularly on the black molds, *Aspergillus niger* van Tiegh and *Rhizopus nigricans* Ehr.[19].

So, the aim of this study to reduce sunflower seed deterioration, when stored under different concentration of two evaporation matters such as propionic and acetic acids both at 25, 50 and 100% throughout 6 months at 2 months intervals. Also, to study the role of both evaporation matters on seedling morphological and physiological characters of sunflower after a period of 6 months from storage under greenhouse conditions.

MATERIALS AND METHODS

A storage experiment was conducted at Seed Technology Research Unit Laboratory in Mansoura, Dakahlia Governorate, Seed Technology Research Department, Field Crop Research Institute, Agricultural Research Center, Egypt. The experiment lasted from November, 2013 to May, 2014. The aim was to

reduce sunflower seed deterioration under different storage periods (0, 2, 4 and 6 months) using different concentrations of two evaporation matters (propionic and acetic acids both at 25, 50 and 100%). In addition, a pot experiment was carried out for 21 days during the summer season of 2014 at greenhouse of the Plant Pathology Department, Faculty of Agriculture, Mansoura University to evaluate the response of sunflower to the tested previous treatments after 6 storage months.

Source of seeds and tested chemical: Sunflower (*Helianthus annuus* L.) Sakha 53 cultivar seeds obtained from the field experiment after harvest were used for storage studies, Oil Crops Research Department, Agricultural Research Center, Egypt. Propionic and acetic acids which were used in this study as a evaporation matters obtained from Al-Gomhoria Company, Egypt.

Seed treatment: In the laboratory, Whatman No.1 filter papers (11 cm diameter) were dipped individually in the evaporation matters (propionic and acetic acids) at various concentrations of 25, 50 and100% until saturation, then two saturated filter papers from each concentration were placed in a separate container (cloth bags) containing the 500 g seeds. Untreated seeds served as control. After evaporation process, the seeds were stored and kept under laboratory conditions for 0, 2, 4 and 6 months. The moisture content of sunflower seeds was 9%.

Laboratory experiments

Seed Health Testing (SHT): Detection of seed-borne mycoflora was carried out following the procedures published by the International Seed Testing Association[20]. Two hundred seeds from each sample and the control were tested using the standard blotter. Ten seeds were plated in 11 cm diameter petri-dish containing three layers of water-soaked blotters using sterilized tap water. The plates were incubated at 20±2°C for 7 days under 12 h alternating cycles of cool white fluorescent light and darkness. Plates were examined under a stereoscopic binocular microscope (6-50 X) for the presence of seed-borne fungi and to study their habit characters. When necessary, the compound microscope was used for confirming the identification after having examined the morphology of conidia and conidiophores. Fungi presented on seeds were identified by means of comparison with the description sheets of Commonwealth Mycological Institute, Kew, Surrey, England (CMI), Danish Government Institute of Seed Pathology (DGISP) publications as well as publications of Raper and Fennell[21], Ellis[22], Chidambaram *et al.*[23], Moubasher *et al.*[24], Booth[25], Burrges *et al.*[26] and Singh *et al.*[27].

Disease assessment: The dead and rotted seeds percentages (non-germinated seed) as well as rotted and survival seedlings percentages were recorded for each storage period.

Germination test: Treated and untreated seeds were subjected to standard germination test as the rules of International Seed Testing Association[28]. Counts of germinating seeds were taken daily up to 10 days after the start of germination. Germination Energy (GE) was recorded as the percentage of germinating seeds at 4th days after plantation is relative to the total number of seeds tested[29].

Germination percentage and seedling characters: Treated and untreated seeds were sown in sterilized sand with the same previous method to determine germination percentage and the seedling characters after 10 days. Germination percentage was defined as the total number of normal seedlings at the end of the test. Seedling length (cm) was determined from 10 normal seedlings and then dried in a forced air oven at 105°C for 24 h to obtain seedlings dry weight (g) under laboratory conditions. Seedling Vigor Index (SVI) was calculated using the equation suggested by ISTA[28]:

$$SVI = \text{Seedling length (cm)} \times \text{Germination percentage}$$

Seed chemical analysis: Seed samples collected periodically from each treatment were oven dried, ground finely for chemical analysis. Seed oil percentage was determined after extraction with Soxhelt's apparatus using petroleum hexane as an organic solvent according to AOAC[30]. Seed nitrogen percentage was estimated by using micro Kjeldahel apparatus and multiplied by the converting factor (6.25) to get seed protein percentage[31].

Greenhouse experiment: Samples of each treatment, as well as untreated control seeds were taking after 6 months storage period and sown in plastic pots for 21 days to study the role of both evaporation matters on seedling morphological and physiological characters.

Morphological characters: Samples of each treatment were cultured in plastic pots (25 cm diameter) which were filled with 2 kg soil mixture (2 sand: 1 clay). Ten sunflower seeds

were sown in 3rd May, 2014 at 21.3°C and relative humidity 58% in each pot. After germination (10 days), the plants were thinned to leave only three seedlings. The experiment was watered using equal amount of water per pot each time. At the end of the experiment (after 21 days from planting), shoot and root lengths (cm), fresh weights of shoot and root (g) and dry weight of shoot and root (g) were measured.

Physiological characters: At 21 days from planting, the blade of the third leaf from the tip was taken to determine photosynthetic pigments (chlorophyll a, b and carotenoids) which were extracted with methanol after adding traces of sodium carbonate[32] and determined according to Mackinney[33]. Total phenols were determined in fresh shoot using the folin- ciocalteau reagent according to Singleton and Rossi Jr.[34].

Statistical analysis: Statistical analysis was performed using analysis of variance technique by means of "MSTAT-C" computer software package for the factorial completely randomized design (Laboratory experiment) and randomized complete block design for greenhouse experiment as published by Gomez and Gomez[35]. Using the Least Significant Difference (LSD) test compared differences among treatment means at the levels of 5% probability.

RESULTS

Laboratory experiments

Seed health testing: Mycological survey of sunflower seed samples showed that 15th species of 11th principal genera of filamentous fungi as shown in Table 1. These genera were *Alternaria, Aspergillus*, *Cephalosporium*, *Fusarium*, *Macrophomina*, *Penicillium*, *Rhizoctonia*, *Rhizopus*, *Verticillium*, *Trichothecium* and *Stemphylium*. The fungal population of some genera i.e., *Alternaria*, *Aspergillus*, *Penicillium*, *Rhizopus* and *Trichothecium* increased with increasing storage period while, other genera decreased in the untreated control. Generally, *Fusarium* was recorded the high frequency followed by *Aspergillus* and *Penicillium*. On contrast, *Rhizoctonia* came late and before it *Stemphlium* and *Cephalosporium*. With regard to species, *Fusarium solani* followed by *Penicillium* sp. then *Aspergillus flavus* and *Rhizopus* sp. recorded the highest frequency in all storage periods. Both evaporations matters (propionic and acetic acids) causes inhibitory effects on the presence of different genera of fungi in all storage periods. This effect increased with increasing concentration of both evaporations from 25-100%.

Generally, acetic acid was recorded the maximum inhibition in fungal populations. The high percentage (100%) was more effective in his respect expect for *Aspergillus ocracius* hence, the high concentrate of propionic acid (100%) was more effective.

Disease assessment: Disease assessment was recorded as dead seeds and rotted seeds (non-germinated seeds) and rotted seedlings, as well as survival seedlings in each storage period. Table 2 shows that dead and rotted seeds as well as rotted seedlings significantly increased with increasing storage periods. Hence, survival seedlings gave inverse this. The highest percentage of dead and rotted seeds as well as rotted seedlings, the lowest percentage of healthy seedlings occurred under untreated control at 6 storage months. Both propionic and acetic acids at any concentration decreased dead and rotted seeds and rotted seedlings but increased healthy survival seedlings under all storage periods. These effects increased with increasing concentration of both evaporation matter. Acetic acid was most effective in this respect.

Germination characters: As for the effect of storage periods, Table 3 shows that germination percentage and its energy decreased significantly with increasing storage period from 0-6 months. Treating sunflower seeds with evaporation matters (propionic and acetic acids) significantly increased germination percentage and germination energy. Propionic acid at 100% followed by acetic acid at 100% gave the highest values of germination characters while, the interaction between storage periods and evaporation matters had no significant effect in this respect.

Seedling characters: Table 4 shows the negative correlation between storage periods and seedlings characters i.e., seedling length, dry weight of 10 seedlings and seedling vigor index. Seed treatment by any concentration of both evaporation matters increased significantly the above parameters as compared with untreated control. In this respect, propionic acid was more effective. The application of evaporation matters decreased the harmful effect of storage periods on seedlings dry weight. Meanwhile, the interaction between storage periods and treatments had no significant effect on seedlings length and seedlings vigor index.

Seed chemical analysis: Table 5 clearly shows that oil and protein percentages in sunflower seeds were decreased significantly with increasing storage periods. The lowest values of these characters resulted from 6 month stored seeds. On

the other side, seed treated with any concentration of the tested evaporation matters increased seed quality (oil and protein percentage) of sunflower at 2, 4 and 6 months storage periods. Propionic acid was the most effective. The interaction

Table 1: Frequency (%) of seed-borne fungal population as affected by two evaporation matters treated sunflower seeds at different storage periods and their interactions

	Alternaria alternate					*Aspergillus flavus*					*Aspergillus niger*				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	12.08	12.98	13.74	15.65	13.61	26.40	44.09	50.61	53.05	43.54	18.92	31.59	32.56	34.37	29.36
Propionic acid (%)															
25	9.89	0.00	2.96	0.00	3.21	21.61	36.09	26.67	36.66	30.26	15.48	25.86	19.20	26.39	21.73
50	8.53	0.00	1.51	0.00	2.51	18.64	31.12	15.77	25.13	22.66	13.35	22.30	11.50	18.32	16.37
100	7.55	0.00	0.00	0.00	1.88	16.51	27.56	6.58	10.48	15.28	1.82	19.75	5.06	8.07	11.17
Acetic acid (%)															
25	8.71	2.96	0.00	0.00	2.91	19.02	31.76	19.33	20.67	22.69	13.63	22.76	14.13	15.20	16.43
50	7.47	1.51	0.00	0.00	2.24	16.32	27.26	11.36	13.01	16.98	11.69	19.53	8.48	9.83	12.38
100	6.32	0.00	0.00	0.00	1.58	16.41	27.40	3.62	0.00	11.85	11.75	19.63	2.99	0.00	8.59
Means	8.65	2.49	2.60	2.23		19.27	32.18	19.13	22.71		13.81	23.06	13.42	16.02	
LSD for															
Storage periods at 5%			0.29					1.17					0.82		
Evaporation matters at 5%			0.38					1.55					1.09		
Interaction at 5%			0.76					3.10					2.17		

	Asperigillus ocracius					*Cephalosporin* sp.					*Fusarium moniliforme*				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	5.84	9.75	10.05	10.61	9.06	8.10	6.92	5.80	5.70	6.63	29.81	28.16	27.26	22.45	26.92
Propionic acid (%)															
25	4.78	7.98	5.93	8.15	6.71	6.63	3.84	4.99	4.97	5.11	24.40	13.67	18.23	17.81	18.53
50	4.12	6.88	2.46	5.65	4.78	5.72	2.43	3.09	0.00	2.81	21.04	8.40	11.08	0.00	10.13
100	3.65	6.10	0.00	0.00	2.43	5.06	1.07	1.56	0.00	1.92	18.64	3.29	5.18	0.00	6.77
Acetic acid (%)															
25	4.21	7.03	4.36	4.69	5.07	5.84	4.99	3.47	0.00	3.57	21.48	18.23	12.25	0.00	12.99
50	3.61	6.03	2.61	3.04	3.82	5.01	3.09	2.12	0.00	2.55	18.43	11.08	0.00	0.00	7.38
100	3.63	6.06	0.92	0.00	2.65	5.04	1.56	0.00	0.00	1.65	18.53	5.18	0.00	0.00	5.92
Means	4.26	7.12	3.76	4.59		5.91	3.41	3.00	1.52		21.76	12.57	10.57	5.75	
LSD for															
Storage periods at 5%			0.35					0.19					0.71		
Evaporation matters at 5%			0.47					0.25					0.93		
Interaction at 5%			0.93					0.50					1.87		

	Fusarium oxysporum					*Fusarium solani*					*Macrophomina phaseolina*				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	47.55	45.15	33.80	23.57	37.52	59.04	54.03	48.45	29.78	47.82	10.30	9.73	9.42	7.76	9.30
Propionic acid (%)															
25	38.92	21.75	29.05	20.70	27.60	48.32	27.39	37.39	25.69	34.70	8.43	4.72	6.30	6.16	6.40
50	33.56	13.33	17.63	18.78	20.82	41.67	16.34	22.30	23.06	25.84	7.27	2.90	3.83	0.00	3.50
100	29.72	5.17	8.20	0.00	10.77	36.90	5.60	9.67	0.00	13.04	6.44	1.13	1.79	0.00	2.34
Acetic acid (%)															
25	34.26	29.05	0.00	16.48	19.94	42.53	37.39	24.34	19.99	31.06	6.42	6.30	4.23	0.00	4.49
50	29.39	17.63	0.00	0.00	11.75	36.50	22.30	0.00	0.00	14.70	6.37	3.83	0.00	0.00	2.55
100	29.54	8.20	0.00	0.00	9.43	36.69	9.67	0.00	0.00	11.59	6.40	1.79	0.00	0.00	2.05
Means	34.71	20.04	12.67	11.36		43.09	24.67	20.30	14.07		7.52	4.34	3.65	1.99	
LSD for															
Storage periods at 5%			1.07					1.36					0.24		
Evaporation matters at 5%			1.42					1.81					0.32		
Interaction at 5%			2.84					3.61					0.65		

Table 1: Continue

	Penicillium sp.					*Rhizoctonia solani*					*Rhizopus* sp.				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	41.86	42.63	44.94	48.24	44.41	6.11	1.86	1.17	0.81	2.48	30.62	35.41	42.28	47.17	38.87
Propionic acid (%)															
25	34.27	18.84	25.26	24.57	25.73	5.00	0.00	0.00	0.00	1.25	25.07	12.77	18.32	16.91	18.27
50	29.54	11.50	15.28	16.21	18.13	4.31	0.00	0.00	0.00	1.07	21.62	7.10	10.50	10.07	12.32
100	26.17	4.38	7.03	0.00	9.39	3.81	0.00	1.17	0.00	0.95	19.14	1.59	3.76	1.41	6.48
Acetic acid (%)															
25	30.15	25.26	16.84	0.00	18.06	4.40	0.00	0.00	0.00	1.10	22.06	18.32	11.14	0.00	12.88
50	25.88	15.28	9.80	0.00	12.74	3.77	0.00	0.00	0.00	0.94	18.93	10.50	5.71	0.00	8.78
100	26.01	7.03	0.00	0.00	8.26	3.79	0.00	0.00	0.00	0.94	19.03	3.76	1.17	0.00	5.99
Means	30.55	17.84	17.02	12.71		4.45	0.26	0.16	0.11		22.35	12.78	13.27	10.79	
LSD for															
Storage periods at 5%			1.07					0.10					0.86		
Evaporation matters at 5%			1.42					0.13					1.13		
Interaction at 5%			2.83					0.27					2.27		

	Verticillium sp.					*Trichothecium* sp.					*Stemphylium* sp.				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	8.74	7.54	6.45	6.36	7.27	25.93	29.98	35.80	39.94	32.91	6.84	5.91	5.05	4.98	5.69
Propionic acid (%)															
25	7.15	4.39	5.28	5.63	5.61	21.23	10.81	15.51	14.32	15.47	5.60	3.44	4.13	4.41	4.39
50	6.16	2.92	3.51	0.00	3.15	18.30	6.01	8.89	8.53	10.43	4.83	2.29	2.75	0.00	2.46
100	5.46	1.51	2.00	0.00	2.24	16.21	1.35	3.18	1.20	5.48	4.27	1.18	1.56	0.00	1.75
Acetic acid (%)															
25	6.29	5.28	4.03	0.00	3.90	18.68	15.51	9.43	0.00	10.90	4.93	4.13	3.16	0.00	3.05
50	5.40	3.51	0.00	0.00	2.23	16.03	8.89	4.83	0.00	7.44	4.23	2.75	0.00	0.00	1.74
100	5.43	2.00	0.00	0.00	1.85	16.11	3.18	0.99	0.00	5.07	4.25	1.56	0.00	0.00	1.45
Means	6.38	3.88	3.04	1.71		18.93	10.82	11.23	9.14		4.99	3.03	2.38	1.34	
LSD for															
Storage periods at 5%			0.20					0.73					0.16		
Evaporation matters at 5%			0.27					0.96					0.21		
Interaction at 5%			0.54					1.92					0.42		

Table 2: Dead seeds, rotted seeds and rotted seedlings as affected by two evaporation matters treated sunflower seeds at different storage periods and their interactions

	Dead seeds					Rotted seeds					Rotted seedlings				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	6.71	6.81	7.96	9.20	7.67	8.06	8.17	9.56	11.06	9.21	4.20	4.73	5.74	12.00	6.66
Propionic acid (%)															
25	4.63	5.57	5.95	7.53	5.92	5.56	6.68	7.14	9.06	7.11	3.13	4.22	5.78	7.40	5.13
50	1.86	3.09	3.71	6.50	3.79	2.65	3.70	4.45	7.80	4.65	2.70	3.63	4.99	6.36	4.42
100	0.96	1.59	2.11	5.73	2.59	1.36	1.91	2.53	6.93	3.18	2.39	3.22	4.41	5.63	3.91
Acetic acid (%)															
25	2.56	4.26	5.57	6.63	4.75	3.65	5.11	6.68	8.00	5.86	2.75	3.71	5.08	6.50	4.51
50	1.63	2.71	3.71	5.73	3.44	2.62	3.67	4.45	6.83	4.39	2.36	3.19	4.37	5.60	3.88
100	0.93	1.54	2.11	5.73	2.57	1.49	2.08	2.53	6.86	3.24	2.37	3.20	4.38	5.63	3.89
Means	2.75	3.65	4.44	6.72		3.63	4.47	5.33	8.08		2.84	3.70	4.96	7.01	
LSD for															
Storage periods at 5%			0.22					0.26					0.20		
Evaporation matters at 5%			0.29					0.35					0.27		
Interaction at 5%			0.58					0.70					0.54		

Table 3: Survival seedlings, germination percentage and germination energy of sunflower seeds as affected by the tested two evaporation matters at different storage periods and their interactions

	Survival seedlings					Germination percentage					Germination energy				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	81.02	80.28	76.73	67.67	76.42	74.00	72.00	72.80	66.30	71.27	74.43	73.33	71.66	68.86	72.07
Propionic acid (%)															
25	86.67	83.52	81.12	76.00	81.82	79.00	74.70	73.30	68.00	73.75	83.00	77.76	75.00	70.00	76.44
50	92.79	89.56	86.85	79.30	87.12	82.00	78.70	76.90	73.00	77.65	84.86	82.00	78.33	75.56	80.19
100	95.28	93.27	90.94	81.67	90.29	88.66	86.66	87.70	74.33	84.34	93.43	86.63	90.00	77.66	86.93
Acetic acid (%)															
25	91.03	86.92	82.65	78.88	84.87	76.66	74.70	74.00	68.00	73.34	80.00	75.56	73.33	69.86	74.69
50	93.37	90.43	87.47	81.86	88.28	80.66	76.00	75.00	72.00	75.91	82.30	82.20	76.66	72.23	78.35
100	95.20	93.17	90.97	81.78	90.28	81.66	81.30	79.30	74.00	79.06	91.83	84.43	86.66	77.66	85.15
Means	90.76	88.16	85.25	78.16		80.38	77.72	77.00	70.80		84.26	80.27	78.81	73.12	
LSD for															
Storage periods at 5%			0.67					2.39					2.32		
Evaporation matters at 5%			0.89					3.16					3.07		
Interaction at 5%			1.78					-					-		

Table 4: Seedling length, dry weight of 10-seedlings and seedling vigor index of sunflower seed as affected by the tested two evaporation matters at different storage periods and their interactions

	Seedling length (cm)					Dry weight of 10-seedlings (g)					Seedling Vigor Index (SVI)				
	Month(s)					Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means	0	2	4	6	Means
Control	18.86	14.40	10.97	10.26	13.62	0.668	0.463	0.365	0.363	0.465	13.96	10.38	7.97	6.76	9.77
Propionic acid (%)															
25	20.39	17.70	12.76	11.26	15.53	0.874	0.596	0.463	0.398	0.583	16.09	13.23	9.36	7.63	11.58
50	20.83	18.55	13.46	11.90	16.18	0.995	0.703	0.531	0.416	0.661	17.09	14.59	10.37	8.66	12.68
100	23.87	23.33	14.30	13.75	18.81	1.203	0.844	0.569	0.435	0.763	21.16	20.23	12.53	10.23	16.04
Acetic acid (%)															
25	18.05	15.90	13.94	10.98	14.72	0.763	0.671	0.433	0.433	0.575	13.92	11.89	10.35	7.46	10.90
50	21.57	19.23	14.66	13.43	17.22	0.887	0.717	0.492	0.453	0.637	17.47	14.54	11.00	9.65	13.17
100	23.26	21.43	15.28	14.33	18.57	0.984	0.728	0.516	0.482	0.677	18.85	17.43	12.09	10.61	14.74
Means	20.98	18.65	13.62	12.27		0.910	0.675	0.481	0.426		16.93	14.61	10.52	8.71	
LSD for															
Storage periods at 5%			1.15					0.056					0.96		
Evaporation matters at 5%			1.52					0.075					1.26		
Interaction at 5%			-					0.150					-		

between storage periods and any of treatments had no significant effect on seed chemical analysis.

Greenhouse experiment

Morphological characters of seedlings: Table 6 shows that the application of both propionic and acetic acid at any dose used increased significantly seedlings morphological character i.e., shoot and root lengths, fresh and dry weights of shoot and root as compared with untreated control seed at 21 days from planting. There is a positive relationship between the concentration of tested materials and their effects. The high concentrate of propionic acid (100%) gives the highest values of seedling morphological characters followed by high acetic acid concentrate (100%).

Physiological characters of seedlings: Seedlings physiological characters were determined as photosynthetic pigments (chlorophyll a, b and carotenoids) and total phenols at 21 days from planting. Table 7 shows that all photosynthetic pigments and total phenols in sunflower seedlings increased significantly by the application of both propionic and acetic acids as compared with untreated control. The low concentrate of acetic acid (25%) give the highest values followed by the high concentrate of propionic acid (100%). It is worthy to mention that the effect of propionic acid was increased by the increase of concentrate. While, the effect of acetic acid was inverse this.

Table 5: Oil and protein percentages of sunflower seeds as affected by the tested agents at different storage periods and their interactions

	Oil (%)					Protein (%)				
	Month(s)					Month(s)				
Treatments	0	2	4	6	Means	0	2	4	6	Means
Control	42.50	38.83	37.70	36.53	38.89	24.33	21.90	18.50	17.19	20.48
Propionic acid (%)										
25	42.16	41.90	38.23	38.05	40.08	23.16	21.30	19.20	19.18	20.71
50	41.90	41.31	38.95	38.07	40.05	23.01	22.50	20.53	19.22	21.31
100	41.60	42.19	39.10	38.72	40.40	24.86	24.80	22.86	21.90	23.60
Acetic acid (%)										
25	42.60	39.09	37.77	37.50	39.24	23.26	22.00	18.80	17.32	20.34
50	40.00	39.37	38.01	37.95	38.83	22.86	22.50	19.50	18.11	20.74
100	39.43	40.10	38.43	38.27	39.06	23.86	23.50	21.65	20.50	22.37
Means	41.45	40.39	38.31	37.87		23.62	22.64	20.15	19.06	
LSD for										
Storage periods at 5%			0.79					0.63		
Evaporation matters at 5%			1.04					0.83		
Interaction at 5%			-					-		

Table 6: Morphological characters of sunflower seedlings as affected by the tested two evaporation matters under greenhouse conditions

Treatments	Shoot length (cm)	Root length (cm)	Shoot fresh weight (g)	Root fresh weight (g)	Shoot dry weight (g)	Root dry weight (g)
Control	21.90	8.63	5.23	0.475	1.159	0.149
Propionic acid (%)						
25	30.20	10.06	7.23	0.608	1.538	0.169
50	32.40	10.20	10.96	0.643	2.212	0.213
100	34.00	12.43	15.80	1.478	2.967	0.420
Acetic acid (%)						
25	27.73	9.23	7.70	0.647	1.330	0.179
50	29.33	10.46	8.60	0.822	1.645	0.209
100	32.40	12.00	12.76	1.420	2.272	0.411
LSD at 5%	3.76	2.90	2.60	0.312	0.494	0.076

Table 7: Physiological characters of sunflower seedlings as affected by the tested two evaporation matters under greenhouse conditions

Treatments	Chlorophyll a (mg g^{-1} fresh weight)	Chlorophyll b (mg g^{-1} fresh weight)	Carotenoids (mg g^{-1} fresh weight)	Total phenols (mg catechol/100 g fresh weight)
Control	1.544	1.334	0.130	136.0
Propionic acid (%)				
25	1.589	1.484	0.171	136.7
50	1.964	1.664	0.288	194.2
100	2.159	1.904	0.493	213.3
Acetic acid (%)				
25	2.324	2.143	0.568	218.2
50	1.664	1.484	0.171	182.8
100	1.934	1.649	0.281	205.6
LSD at 5%	0.087	0.081	0.049	10.1

DISCUSSION

The present study shows that the deterioration of sunflower stored seeds increased with increasing storage period from 0-6 months. On the other side, seed treatment with propionic and acetic acids as evaporation matters decreased the presence of fungal genera on sunflower seeds under storage conditions up to 6 months. Also, inhibited disease development by decreasing dead and rooted seeds as well as rooted seedlings. In addition to the enhancement in germination characters and seed chemical components as well as morphological and physiological characters of seedlings. Sunflower seeds have limited longevity due to its chemical composition from oil and fatty acids. The main reasons for the rapid deterioration of oily seeds are autoxidation of lipids and increase in the content of free fatty acids[5]. Seed deterioration during storage beginning with declines of seed vigor followed by loss of germination and viability[36]. The usage of lipids in the respiration process leading to significant reduction of oil content in seed[6]. They stated that the types of fatty acids present in an oil and in particular their number of double bonds determine the type

and extent of chemical reactions which occur during the storage time. The product resulting from lipid peroxidation lead to DNA denaturation, prevent translation and protein transcription and cause oxidation of the most reactive amino acids which decrease in vigor and seed germination[37,38]. Also, Verma *et al.*[39] stated that the dehydrogenase activity was reduced as the aging progressed and was lowest after four years of storage in *Brassica* spp.[10] revealed that seed oil concentration decreased with time regardless of storage conditions. In addition to the increase of seed deterioration during storage may be due to increasing of metabolic activity such as oxidation of amino acids and respiratory activity which decreases the reserve substances content and advance in the deterioration of stored seeds[40]. The metabolism of seeds during storage to provide energy for its physiological activity lead to reduction in seed oil[41]. It is stated that the main enzymes which are involved in oil and fatty acids metabolism of the seeds are acyl-CoA oxidase, malate synthase, ctrate synthase, catalase and lipases.

The storage fungi are considered one of the most reasons of seed deteriorations within storage period. Its causes decrease viability, discoloration, produce mycotoxin, heat production and develop mustiness and caking[40]. Moreover, Magan *et al.*[42] classified the factors which lead to seed deterioration by storage fungi into four groups. These groups are namely: Intrinsic nutritional factors extrinsic factors, processing factors and implicit factors, which affect the fungal population that could change throughout the storage period.

Propionic and acetic acids have been thoroughly proved to be the antimicrobial food preservatives and acidulants[19]. Fumigation with acetic, formic and propionic acids are reported to prevent decay of cherries, stone fruit, pome and citrus fruit by destroying spores of four different pathogens on the fruit surface[43]. It was shown that vapor of acetic and propionic acids would kill spores of pathogenic fungi on the surface of fruits[44,45]. The use of propionic acid and propionates has been directed primarily against molds, although some yeasts and bacteria, particularly Gram-negative strains may also be inhibited[18].

CONCLUSION

The obtained results in this study showed that propionic and acetic acids vapors can have a considerable fungicidal activity against sunflower pathogens and improve seed viability. Generally, it could be concluded that using 100% propionic acid as sunflower seed treatment tended to reduce the deterioration and seed-borne pathogens of sunflower under storage conditions.

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