J. Agric. Sci. Mansoura Univ., 28(12): 8053 - 8061, 2003

ANTIFUNGAL ACTIVITY OF SOME ORGANIC ACIDS USED AS FEED PRESERVATIVES

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ABSTRACT

Organic acids (formic, propionic, acetic, lactic, citric and fumaric acids) were used in different concentrations (25%, 35%, 45%, 55%, 65%, 75% and 100%) at different inclusion rates (0.1%, 0.2%, 0.4%, 0.6%, 0.8%, 1.0%, 1.2%, 1.4% and 1.6%) for all acids except acetic (0.05%, 0.1%, 0.2%, 0.4%, 0.6%, 0.8%, 1.0%, 1.2% and 1.4%) and formic and propionic acids (0.02%, 0.025%, 0.05%, 0.1%, 0.15%, 0.2%, 0.4% and 0.6%) to evaluate their antimould activity to be used as grain preservatives. Results showed that, propionic acid (100%) has the strongest effect as antimould at 0.02% inclusion rate followed by formic and acetic acids (100%) at inclusion rate 0.05% for both of them. Diluted acids has effective antimould activity but in higher inclusion rates. Citric acid, lactic acid and fumaric acid did not have any antifungal effect up to 1.6% inclusion rate but they showed an enhancement effect on the fungal growth.

Keywords: Organic acids, antimould activity, propionic, acetic, formic, citric, lactic, fumaric, preservatives, corn.

INTRODUCTION

Corn grain is often harvested at a moisture content which may enhance the growth, colonization and mycotoxins production by a range of fungi. If drying to safe moisture content (14-15%) is delayed or inefficient, fungi can rapidly colonize the grain causing harmful effects which vary depending on relative humidity and storage temperature. So, preservatives are used in treating such grains for controlling both spoilage and mycotoxins producing fungi during storage (Marine *et al.*, 2002).

Chemical preservatives have been defined by Food and Drug Administration as "any chemical that when added to food tends to prevent or retard deterioration". There are approximately 30 different compounds which can legally be used as antimicrobials in food/feed products (Fulton, 1980). The mechanism of action of preservative is usually based on either: 1) destruction of cell wall or cell membrane, 2) inhibition of various enzymes in the microbial cell or 3) destruction of the genetic structure of the protoplasm (Robert *et al.*, 1995).

Selection of the proper preservative is dependant upon several factors: a) Antimicrobial spectrum. b) Antimicrobial activity. C) Chemical and physical properties. D) Relative toxicity. E) Resistance development. F) Organoleptic properties. G) Economical consideration and I) A suitable procedure for analysis (Robert *et al.*, 1995).

Organic acids are one of the most important preservatives which are permitted in many countries and can be divided into two groups: One group shows an antimicrobial activity by reducing the pH and includes: acetic acid, citric acid, formic acid and lactic acid. Several reports have shown that, also the undissociated form of acetic acid has antimicrobial action (Gulam *et al.*, 1987, Robert *et al.*, 1995). Also fumaric acid has an antimicrobial effect by reducing the pH of the substrate (Voget *et al.*, 1981). The other group of organic acid preservatives, including propionic acid, show an antimicrobial activity only when they are present as undissociated acids. However, the dissociated form of these acids may have an antimicrobial effect (Skirdel and Eklund, 1993).

The use of sub-optimal levels of organic acids results in a significant enhancement of the fungal count and mycotoxins production as the mycoflora of the treated substrate utilize these acids as nutritive substances (Marine *et al.*, 2002).

The objectives of the present work were to investigate the effectiveness of acetic, citric, formic, fumaric, lactic and propionic acids as preservatives and to determine their effective concentrations as antifungal substances.

MATERIALS AND METHODS

Preparation of different concentrations of examined organic acids:

Different concentrations of the examined organic acids were prepared using autoclaved distilled water as a diluent in case of formic, propionic, acetic and lactic acids and autoclaved finely grounded corn as a diluent in case of citric and fumaric acids. The prepared concentrations were 25%, 35%, 45%, 55%, 65%, 75% and 100%.

Determination and adjustment of the moisture content of corn before inoculation:

Moisture content of corn sample was estimated according to (AOAC, 1998) then the moisture content was adjusted to be 18% according to the following equation:

$$S = \frac{\text{Required moisture content} - \text{Initial moisture content}}{100 - \text{Required moisture content}} \times 100$$

Where S = the volume of water required for 100 gm of sample to reach the required level of moisture content.

Experimental design:

One kg of corn 18% moisture content was used for each concentration. This amount was equally divided into ten sterile Erlenmeyer flasks; each flask contained 100 gm of corn. The prepared previously mentioned concentrations of organic acids included in this study were added to the corn subsamples with different inclusion rates (0.1%, 0.2%, 0.4%, 0.6%, 0.8%, 1.0%, 1.2%, 1.4% and 1.6%) for all acids except acetic (0.05%, 0.1%, 0.2%, 0.4%, 0.6%, 0.8%, 1.0%, 1.2% and 1.4%) and formic and propionic acids (0.02%, 0.025%, 0.05%, 0.1%, 0.15%, 0.2%, 0.4% and

0.6%). One flask was left without inoculation as control. Ten grams were withdrawn from each flask just after inoculation with the acids (zero time), after 7 days and neht after 14 days to estimate the changes in the fungal content. The Total Fungal Count was performed as follows: Ten grams of each sample were added to a 90 ml sterile saline solution in 500 ml Erlenmeyer flasks and homogenized thoroughly on an electric shaker at a constant speed. Tenfold serial dilutions were then prepared. One ml portion of three suitable dilutions of the resulting sample suspension were used to inoculate Petri dishes each containing 15 ml Sabaroud Dextrose Agar containing 0.5 mg Chloramphenicol/ml medium to inhibit bacterial growth. Plates were then incubated for 7 days at 28°C and the grown fungal colonies were counted (Aziz *et al.*, 1998).

Criteria of acceptance:

The criteria of evaluation of antifungal activity are estimated according to European pharmacopoeia (2001) in terms of the log reduction in the number of viable microorganisms against the value obtained for the fungal count at zero time.

RESULTS AND DISCUSSION

This work has shown the interaction between fungal colonization and preservatives in the stored grains ecosystem.

It is clear from all tables that, the total fungal count increased 1 log during the whole period of the experiment in the control group which received no treatments. This result is supported by the findings of Philip *et al.*, 1983 and Marine *et al.* 2002 who stated that in the absence of preservatives, fungal growth is fast and obvious.

Table (1) shows the fungal behavior in corn treated with different concentrations of acetic acid included in various rates. It is clear that, at the concentrations 25%, 35%, 45%, 55%, and 65% the effective inclusion rate was 0.2% which was indicated by reduction of the log number by 1 after two weeks. At 75% the effective rate was 0.1% and at 100%, the effective rate was 0.05%. This result is similar to that of Gulam *et al.* 1987 who found an obvious antimould activity of acetic acid when used against different types of fungi which was indicated by the reduction of total fungal count.

Data in Table (2) shows the effect of citric acid on fungal content of corn with 18% moisture content. It is clear that there was no inhibitory effect of any of the used concentrations at any inclusion rate on the fungal count. All of the used concentrations had an enhancement effect on the fungi which was indicated by the increase of the log number by 1 or 2. Conkova *et al.*, 1993 also proved that up to 5% of citric acid had no antifungal effect on microscopic fungi.

Table (1): Antifungal activity of different concentrations of Acetic acid at different inclusion rates at 18% moisture content:

Inclusion rate (%)	Duration / Day	noitulid fo rewoP						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
0.05	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	102
0.1	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	102	102
0.2	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
0.4	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
0.6	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
0.8	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
1.0	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	102	10^{2}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
1.2	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{2}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
1.4	0 (Before inoculation)	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
	7	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{2}
	14	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}

Table (2): Antifungal activity of different concentrations of Citric acid at different inclusion rates in corn of 18% moisture content:

Inclusion rate (%)	Duration / Day	noitulid fo rewoP						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10	10	10	10	10	10	10
	7	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
0.1	0 (Before inoculation)	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10	10
	7	10^{2}	10^{2}	10^{2}	10^{3}	10^{4}	10^{2}	10^{2}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{3}
0.2	0 (Before inoculation)	10	10	10^{2}	10^{2}	10	10	10^{2}
	7	10^{3}	10^{2}	10^{2}	10^{3}	10^{2}	10	10^{2}
	14	10^{4}	10^{3}	10^{3}	10^{3}	10^{2}	10^{2}	10^{3}
0.4	0 (Before inoculation)	10^{2}	10	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}
	7	10^{2}	10^{2}	10^{2}	10^{3}	10^{2}	10^{2}	10^{3}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}
0.6	0 (Before inoculation)	10^{2}	102	10^{2}	10^{2}	10^{2}	10	10^{2}
	7	10^{3}	10^{3}	10^{2}	10^{3}	10^{2}	10^{2}	10^{2}
	14	10^{4}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{3}
0.8	0 (Before inoculation)	10	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10
	7	10^{3}	10^{2}	10^{2}	10^{3}	10^{2}	10^{2}	10^{2}
	14	10^{4}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}
1.0	0 (Before inoculation)	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10^{2}	10
	7	10^{3}	10^{2}	10^{2}	10^{3}	10^{2}	10^{2}	10^{2}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}
1.2	0 (Before inoculation)	10	10^{2}	10^{2}	10^{2}	10^{2}	10	10^{2}
	7	10^{3}	10^{2}	10^{2}	10^{3}	10^{2}	10^{2}	10^{3}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{3}
1.4	0(Before inoculation)	10	10^{2}	10^{2}	10^{2}	10	10^{2}	10
	7	10^{2}	10^{3}	10^{2}	10^{3}	10	10^{2}	10^{2}
	14	10^{4}	10^{3}	10^{3}	10^{3}	10^{2}	10^{3}	10^{2}
1.6	0 (Before inoculation)	10	10^{2}	10^{2}	10^{2}	10	10^{2}	10
	7	10^{3}	10^{2}	10^{2}	10^{3}	10	10^{2}	10^{2}
	14	10^{3}	10^{3}	10^{3}	10^{3}	10^{2}	10^{3}	10^{2}

Table (3) shows the antifungal effect of formic acid. The effective inclusion rate was 0.6% for concentrations 25%, 35% and 45% and was 0.15% at 55%. At 65% and 75%, the effective inclusion rate was 0.1% and at 100% it was 0.05%. Schultz and Muller 1999 found that formic acid was effective as antimould when used in a higher inclusion rates (1-2%) than that used in this study. This result may be due to differences in moisture content, higher dilutions of the acid or due to bad mixing at the beginning of the experiment. Many researchers reported the effectiveness of formic acid as antimould agent like Holemberg *et al.* 1989, Ramos and Hernandez, 1993, Tzatzarakis, 2000.

Table (3): Antifungal activity of different concentrations of Formic acid at different inclusion rates at 18% moisture content:

Inclusion rate (%)	Duration / Day	Power of dilution						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.02	0 (Before inoculation)	10^2	10^2	10	10	10^2	10^2	10
	7	10^2	10^2	10	10	10^2	10^2	10
	14	10^2	10^2	10	10	10^2	10^2	10
0.25	0 (Before inoculation)	10	10^2	10	10	10^2	10	10
	7	10	10^2	10	10	10^2	10	10
	14	10	10^2	10	10	10^2	10	10
0.05	0 (Before inoculation)	10^2	10	10	10	10^2	10	10
	7	10^2	10	10	10	10^2	10	-
	14	10^2	10	10	10	10^2	10	-
0.1	0 (Before inoculation)	10^2	10	10^2	10^2	10^2	10	10
	7	10^2	10	10^2	10^2	10	-	-
	14	10^2	10	10^2	10^2	10	-	-
0.15	0 (Before inoculation)	10^2	10	10^2	10	10^2	10	10
	7	10^2	10	10^2	-	10	-	-
	14	10^2	10	10^2	-	10	-	-
0.2	0 (Before inoculation)	10^2	10^2	10^2	10	10^2	10^2	10
	7	10^2	10^2	10^2	-	10	10	-
	14	10^2	10^2	10^2	-	-	10	-
0.4	0 (Before inoculation)	10	10	10	10	10^2	10^2	10
	7	10	10	10	-	10	10	-
	14	10	10	10	-	-	10	-
0.6	0 (Before inoculation)	10	10	10	10	10^2	10^2	10
	7	-	-	-	-	10	10	-
	14	-	-	-	-	-	-	-

Data in Table (4) indicated that the addition of fumaric acid at any of the used concentrations and inclusion rates had no antifungal effect. Also, the obtained data showed that, the use of suboptimal doses of this acid caused an increase of the total fungal count by 1 to 2 logs. Lactic acid also had the same effect when it was used in the previously mentioned concentrations and inclusion rates (Table 5). El Gazzar *et al.* 1987 found that up to 2% of lactic acid used as antifungal agent. The increase of fungal count which was the result of addition of suboptimal concentration of organic acids was supported by data obtained by Marine *et al.*, 2002 who concluded that lower doses of preservatives enhances fungal growth during storage. Schultz and Muller 1999 reported the failure of lactic acid to act as feed preservative or antifungal agent when used in various concentrations. This result also is supported by Higgins and Brinkhause, 2000 who found that this acid had no effect up to 1%.

Table (4): Antifungal activity of different concentrations of Fumaric acid at different inclusion rates at 18% moisture content:

Inclusion rate (%)	Duration / Day	noitulid fo rewoP						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10	10	10	10	10	10	10
	7	102	102	102	102	102	102	102
	14	103	103	103	103	103	102	103
0.1	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^2
0.2	0 (Before inoculation)	10^2	10^2	10^2	10^2	10	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^2	10^3	10^3
0.4	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^2
0.6	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.8	0 (Before inoculation)	10	10^2	10^2	10^2	10^2	10	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^2	10^3	10^3	10^3	10^3	10^2	10^3
1.0	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
1.2	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
1.4	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^2	10^3
1.6	0 (Before inoculation)	10^2	10^2	10^2	10^2	10	10	10^2
	7	10^2	10^2	10^2	10^2	10	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^2	10^2	10^3

Table (5): Antifungal activity of different concentrations of Lactic acid at different inclusion rates at 18% moisture content:

Inclusion rate (%)	Duration / Day	noitulid fo rewoP						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.1	0 (Before inoculation)	10^2	10^2	10^2	10	10^2	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.2	0 (Before inoculation)	10^2	10	10^2	10^2	10^2	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^2	10^3	10^3	10^3	10^3	10^3
0.4	0 (Before inoculation)	10	10	10^2	10	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.6	0 (Before inoculation)	10^2	10	10^2	10	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.8	0 (Before inoculation)	10^2	10^2	10^2	10	10^2	10	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
1.0	0 (Before inoculation)	10^2	10^2	10^2	10	10^2	10^2	10
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
1.2	0 (Before inoculation)	10^2	10	10	10	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^2	10^3	10^3	10^3	10^3
1.4	0 (Before inoculation)	10^2	10	10^2	10	10	10	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^2	10^3	10^3	10^3
1.6	0 (Before inoculation)	10^2	10	10	10	10^2	10	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^2	10^3	10^3	10^3

Table (6) shows the antimould effect of propionic acid. It is clear from the obtained data that the effective inclusion rates were: 0.1% at 25%, 35% and 45%, 0.05% at 55% acid concentration, 0.025% at 65% and 75% and 0.02% at 100% acid concentration. These data agree with that obtained by Gulam *et al.*, 1987 who reported that propionic acid is the most effective antifungal preservative for corn. The effective dose which is reached in this study (0.02%) agreed with that obtained by Philip *et al.* 1983.

Table (6): Antifungal activity of different concentrations of Propionic acid at different inclusion rates at 18% moisture content:

Inclusion rate (%)	Duration / Day	noitulid fo rewoP						
		25%	35%	45%	55%	65%	75%	100%
0	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^3	10^3	10^3	10^3	10^3	10^3	10^3
0.02	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^2	10^2	10^2	10^2	10^2	10^2	10
0.25	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^2	10^2	10^2	10^2	10	10	10
0.05	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	14	10^2	10^2	10^2	10	10	10	10
0.1	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10
	14	10	10	10	10	10	10	10
0.15	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10^2	10^2	10
	14	10	10	10	10	10	10	10
0.2	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	102	10^2
	7	10^2	10^2	10^2	10^2	10^2	10	10
	14	10	10	10	10	10	10	-
0.4	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	102	10^2
	7	10^2	10^2	10^2	10^2	10	10	-
	14	10	10	10	10	10	10	-
0.6	0 (Before inoculation)	10^2	10^2	10^2	10^2	10^2	10^2	10^2
	7	10^2	10^2	10^2	10^2	10	10	-
	14	10	10	10	10	10	-	-

Data in Table (7) summarize the results obtained allover the time of the experiment. It is clear that all the inclusion rates of the different concentrations of the citric, fumaric and lactic acids had no antimould effect while propionic acid had the strongest antimould effect. At concentrations 25%, 35% and 35%, acetic acid has stronger antimould activity than formic acid while at concentrations 55%, 65% and 75% formic acid was more effective. At 100% concentration (pure acids) both acids had the same antimould effect.

Table (7) Percentage of effective doses of organic acids as antifungal compounds at 18% moisture contents:

Concentrations Acids	25%	35%	45%	55%	65%	75%	100%
Acetic	0.2	0.2	0.2	0.2	0.2	0.1	0.05
Citric	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6
Formic	0.6	0.6	0.6	0.15	0.1	0.1	0.05
Fumaric	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6
Lactic	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6	>1.6
Propionic	0.1	0.1	0.1	0.05	0.025	0.025	0.02

Acetic acid, formic acid and propionic acid caused fixation of the total fungal count when added at lower concentrations and inclusion rates than the inhibitory doses while citric acid, fumaric acid and lactic acid caused enhancement of the fungal growth when used at lower levels (Tables 1-6). Tzatzarakis *et al.* (2000) reported the advantage of formic acid on acetic acid as feed preservative. Culam *et al.* (1987) observed that propionic acid had a stronger antimould effect than acetic acid while Holemberg *et al.* (1989) observed that the same acid (propionic) had stronger effect than formic acid.

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النشاط المضاد للفطريات لبعض الأحماض العضوية التى تستخدم كمـــواد حافظـــة للأعلاف

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المعمل المركزى للأغذية و الأعلاف– مركز البحوث الزراعية

تم إستخدام الأحماض العضوية (الفورميك – الـــبروبيونيك – الخليــك – اللاكتيــك – الستريك – الفيوماريك) بتركيزات مختلفــة (٢٥ % ، ٣٥ % ، ٤٥ % ، ٥٥ % ، ٦٥ % ، ٧٥ % ، ١٠٠ %) و بمعدلات إضافة مختلفــة (٠,١ % ، ٠,٢ % ، ٠,٤ % ، ٠,٦ % ، ٠,٨ % ، ١ % ، ١,٢ % ، ١,٤ % ، ١,٦ %) لكل الأحماض ما عــدا الخليــك (٠,٠٥ % ، ٠,١ % ، ٠,٢ % ، ٠,٤ % ، ٠,٦ % ، ٠,٨ % ، ١ % ، ١,٢ % ، ١,٤ %) ، (٠,٠٢ % ، ٠,٠٢٥ % ، ٠,٠٥ % ، ٠,١ % ، ٠,١٥ % ، ٠,٢ % ، ٠,٤ % ، ٠,٦ %) لحمــض الفورميـــك و البروبيونيك.

أظهرت النتائج أن حمض البروبيونيك له أقوى تأثير كمضاد للفطريات عند تركيز ١٠٠ % بمعدل إضافة ٠,٠٢ % يليه حمض الفورميك و الخليك ١٠٠ % بمعدل إضافــة ٠.٠٥ %. و أظهرت النتائج أيضا أن إستخدام هذه الأحماض بتركيزات أقل له تأثير مضاد للفطريـــات و لكـــن بمعدلات إضافة أكبر من المشار إليها.

لم تظهر باقى الأحماض (الستريك و اللاكتيك و الفيومـــاريك) اى تـــأثيرات مقاومـــة للفطريات بكل التركيزات و معدلات الإضافة المستخدمة و لكن على العكس أظهرت تأثيرا محفزا لنمو الفطريات.