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ANTIBACTERIAL EFFECT OF HERBS AND SPICES USED IN THE PRODUCTION OF SOME DAIRY PRODUCTS

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ABSTRACT

The antibacterial effect of twelve raw herb and spice plants (Rosemary, Thyme, Ginger, Common fennel, Coriander, Black cumin, Black pepper, Cayenne pepper, White pepper, Turmeric, Cinnamon and Red pepper) against the most common eight dairy spoilage and poisoning bacteria (*Bacillus cereus a* ATCC 10987, *Bacillus cereus b* ATCC 14049, *Bacillus subtilis* ATCC 6633, , *Staphylococcus aureus* ATCC 29740, *Yersinia enterocolitica* ATCC 23715, *Salmonella typhimurium* ATCC 2556, *E.coli* O157:H7, *Listeria monocytogenes* ATCC 51778) was examined. The disc assay and agar well diffusion methods on a specific media were used for antibacterial activity determination to four concentrations of raw herb and spices and the zone of inhibition diameter were estimated.

All thyme concentrations had no inhibitory effect on *Bacillus subtilis* and *Yersinia enterocolitica*. While, *E. coli* (O157: H7), *Listeria monocytogenes* and *Staph.aureus* were totally inhibited by the thyme concentration. Common Fennel had some antibacterial effects against *Bacillus cereus*, *E. coli*(O157: H7), *Listeria monocytogenes*, *Salmonella typhimurium* and *Staph.aureus*. While, Common Fennel concentration had no antibacterial effects on *Bacillus subtilis* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was generally found against *Bacillus cereus* and *Staph.aureus*, by increasing the Common Fennel concentration the antibacterial activity increased. On the contrary, Black-Cumin concentration was found to have no antibacterial activity against most of tested dairy spoilage and poisoning bacteria. Coriander had some antibacterial effects against *Bacillus cereus*, *E. coli* (O157: H7),

Listeria monocytogenes, and *Salmonella typhimurium*. While, Coriander concentration had no antibacterial effects on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was found against *Bacillus cereus* and *E. coli*. The inhibition of dairy spoilage and poisoning bacteria increased by increasing the thyme concentration. Also, all black-Pepper concentrations had no inhibitory effect on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. Generally, *Bacillus cereus* and *E. coli* (O157: H7), and were totally inhibited by the black-pepper concentration. In conclusion, Rosemary and Ginger, Thyme and common fennel were found to be the most inhibitory plants and could be recommended on the production dairy products.

Key words: Ginger, Thyme, Rosemary, Common fennel, Antimicrobial, spoilage, poisoning bacteria, Dairy products.

INTRODUCTION

Herbs and spices have been added to foods since ancient times, not only as flavoring agents, but also as folk medicine and food preservatives. In addition to imparting characteristic flavors, certain spices and herbs prolong the shelf life of foods by preventing rancidity throughout their antioxidant activity or through bacteriostatic or bacteriocidal activity. Spices have been defined as plant substances from indigenous or exotic origin, aromatic or with strong taste, used to enhance the taste of foods. Spices include leaves (bay, mint, rosemary, coriander, and oregano), flowers (clove), bulbs (garlic, onion), fruits (cumin, red chilli, and black pepper), stems (coriander, cinnamon), rhizomes (ginger), and other plant parts. Although herbal spices have been well known for their medicinal, preservative and antioxidant properties, they have been currently used with primary purpose of enhancing the flavor of foods rather than extending shelf-life. (Smid, and Gorris 1999; and Stermitz *et al.*, 2000).

There are some few searches on the antibacterial effect of aqueous extracts of herbs and medicinal plants as compared with their oils. All the extracts inhibited the growth of all of the tested organisms which include enterotoxigenic *Escherichia coli* (ETEC), *Shigella dysenteriae*, *Staphylococcus aureus*, *Salmonella typhi*, but the leaf extract in schnapps without *Zingiber officinale* gave the highest

inhibition on the growth of the test organisms with diameter ranging from 7.0-16.0 mm.

Essential oils of commercially available oils of lavender (*Lavandula angustifolia* Mill.), lemon-balm (*Melissa officinalis* L.), juniper berry (*Juniperus communis* L.), lemon verbena (*Lippia citriodora* Kunth), rosemary (*Rosmarinus officinalis* L.) and cypress (*Cupressus sempervirens* L.) had antimicrobial activity, and *E. coli* was the most sensitive organism among the tested ones to the inhibitory effect of the oils (Adebolu *et al.*, 2007 and Romeo *et al.*, 2008).

Garlic, onion, allspice, and oregano were effective seasoning inhibited 100 % to the bacterial species being tested.. Next on the list, several spices inhibiting more than 75% of the microbial species. These were thyme, cinnamon, tarragon, cumin, cloves, lemongrass, bay leaf, capsicums and rosemary. Spices having inhibitory action less than 75% of the microbial species were marjoram, mustard, caraway, mint, sage, fennel, coriander, dill, nutmeg, basil, parsley, cardamom, pepper (white & black), and ginger (Shan *et al.*, 2007).

Out of 46 spices and herb extracts, 12 exhibited high antibacterial activities against five food- borne bacteria (*Bacillus cereus*, *Listeria monocytogenes*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella anatum*) Shan *et al.*, (2007)

Therefore, the work aimed to investigate the antibacterial effect of different concentrations for different herbs and spices on the most common spoilage bacteria in dairy products.

MATERIALS AND METHODS

1. Materials:

Herb and spices:

Twelve raw herb and spice plant types were purchased from Cairo governorate spice Market. These plants are:

Rosemary (*Rosmarinus officinalis* L.), Thyme (*Thymus vulgaris*), Ginger (*Zingiber officinale*), Common fennel (*Foeniculum vulgare*), Coriander (*Coriandrum sativum*), Black cumin (*Nigella sativa*), Black pepper (*Piper nigrum*), Cayenne pepper (*Capsicum annum*), White pepper (*P. guineense*), Turmeric (*Curcuma longa*), Cinnamon (*Cinnamomum cassia*) and Red pepper.

Pathogenic and dairy spoilage bacteria:

Eight different bacterial strains cause the spoilage and poisoning of dairy products were used throughout this work, and these strains are:

Bacillus cereus a (ATCC 10987), *Bacillus subtilis* (ATCC 6633), *Bacillus cereus b* (ATCC 14049), *Staphylococcus aureus* (ATCC 29740), *Yersinia enterocolitica* (ATCC 23715), *Salmonella typhimurium* (ATCC 2556), *E.coli* (O157:H7), *Listeria monocytogenes* (ATCC 51778). The strains were obtained from the Egyptian Microbial Culture Collection (EMCC) at Cairo Microbiological Resources Center (Cairo MIRCEN), Faculty of Agriculture, Ain Shams University.

Media:

The following media were used for cultivation, activation and enumeration of the examined bacteria. Tryptic soya broth and agar media for activation of all bacterial strains. Specific media were used for enumeration of bacteria and antibacterial determination. While, Nutrient agar medium was used for *Bacillus* strains. Baird-parker agar medium (Difco) for *Staph.aureus*. *Yersinia* CIN agar medium (Oxoid) for *Yersinia*, Bismuth sulphite agar medium (Difco) for *Salmonella*. MacConkey agar medium (Difco) for *E.coli*. Palcam agar medium (Difco) for *Listeria monocytogenes* according to the methods described by Marshall (1992).

2. Methods:**Cultivation and activation of bacterial strains:**

Bacterial strains were precultured and activation in tryptic soya broth and agar media. At 32°C for 24 h followed by cooling to 4 ± 1°C till the end of experiments.

Herb and spice plants extract preparation :

Crude herb and spice plants were weighted (100 g / 1000 L) and autoclaved at 121°C for 5 min, followed by cooling and filtered using cheese cloth bags under sterile conditions.

Antibacterial activity:

Antibacterial activity was determined according to Magnusson and Schnurer (2001). Activity was determined by measuring the

diameter of zones (mm) showing complete inhibition. Nutrient agar was used as medium for estimating antibacterial activity used as antibacteriological media. Antibacterial potency of the compound was measured against all used strains for confirming the data according to the standard disc diffusion method. (Magnusson and Schnurer, 2001).

Statistical analysis:

The data were analyzed according to Statistical Analysis System User's Guide (SAS, 1994) (SAS, Institute, Inc, U.S.A.).

RESULTS AND DISCUSSION

Effect of thyme concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm) presented in Table (1) reveal that the inhibition of dairy spoilage and poisoning bacteria increased by increasing the thyme concentration. Also, all thyme concentrations had no inhibitory effect on *Bacillus subtilis* (ATCC 6633) and *Yersinia enterocolitica* (ATCC 23715). Generally, *E. coli* (O157:H7), *Listeria monocytogenes* (ATCC 51778) and *Staph.aureus* (ATCC 29740) were totally inhibited by the thyme concentration.

Table (1): Effect of thyme concentration (% w/vol) on some different spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	1.9±0.10	5.5±0.31	9.0±0.18	10.0±0.22
<i>Bacillus cereus</i> ATCC 10987	2.1±0.12	4.0±0.14	8.9±0.27	9.7±0.18
<i>Bacillus subtilis</i> ATCC6633	0	0	0	0
<i>E. coli</i> (O157 : H7)	9.9±0.42	12.5±0.34	17.8±0.38	20.0±0.50
<i>Listeria monocytogenes</i> ATCC 51778	8.9±0.18	10±0.20	11.6±0.21	13.0±0.45
<i>Salmonella typhimurium</i> ATCC 2556	6.0±0.15	9.1±0.11	10.8±0.24	13.8±0.40
<i>Staph.aureus</i> ATCC 29740	4.2±0.13	5.1±0.10	9.9±0.19	12.6±0.32
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

The data in Table (2) show the effect of Common Fennel concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition by mm). Common Fennel had some antibacterial effects against *Bacillus cereus*, *E. coli* (O157: H7), *Listeria monocytogenes*, *Salmonella typhimurium* and *Staph.aureus*. While, Common Fennel concentration had no antibacterial effects on *Bacillus subtilis* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was detected against *Bacillus cereus* and *Staph.aureus*. Generally, by increasing the Common Fennel concentration, the antibacterial activity increased.

Kamble and Patil (2007) evaluated the aqueous extracts of 32 spices for antimicrobial activities against 15 species of bacteria and 9 species of yeasts. Garlic exhibited the greatest antibacterial activity, followed by clove. Star anise (*Illicium verum*), tamarind, cinnamon (*Cinnamomum zeylanicum*) and ginger showed moderate inhibitory activities. On the contrary, the lowest inhibitory activities were obtained with extracts of turmeric, asafoetida (*Ferula assa-foetida*), cardamom (small and large), cumin (*Cuminum cyminum*), basil (*Ocimum basilicum*), parsley, mustard (*Brassica juncea*) and nutmeg (*Myristica fragrans*), inhibiting the growth of 1-2 bacteria only. Garlic, cinnamon and clove were the most inhibitory spices against yeasts, while ginger, asafoetida, mustard, mace (*Myristica fragrans*), black pepper (*Piper nigrum*), cardamom (large), curry leaf (*Murraya koenigii*) and dill (*Anethum graveolens*) exhibited moderate inhibitory activities. Caraway (*Carum carvi*), fenugreek, cumin, fennel, coriander (*Coriandrum sativum*), aniseed (*Pimpinella anisum*), mint (*Mentha piperita*), star-anise, parsley, nutmeg, tamarind, turmeric, poppy seed (*Papaver somniferum*), ajowan (*Trachyspermum ammi*), rosemary and pomegranate were the least inhibitory. Kokam (*Garcinia indica*), pepper long (*Piper longum*), basil, cardamom (small) and saffron had no inhibitory effect against the yeasts.

Table (3) presents the effect of black-cumin concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). Black-Cumin concentration was found to have no antibacterial activity against most of tested dairy spoilage and poisoning bacteria. Black-Cumin concentration could be only effective in inhibition of *Staph.aureus* and *Bacillus subtilis*, and the antibacterial activity increased with increasing of black-Cumin concentration. Shah and Ray (2003) reported that Gram positive

organisms (*Staphylococcus aureus*, *S. aureus* (ATCC), *Bacillus cereus*, and *Streptococcus faecalis* [*Enterococcus faecalis*]) were more sensitive to black cumin raw seed powder as compared to Gram negative organisms (*Escherichia coli*, *E. coli* (ATCC), *Pseudomonas aeruginosa*, and *Salmonella typhi*). Also, moisture free black cumin seed powder was more effective in its antimicrobial activity than original seed powder. The probable active principles present in different extracts of black cumin seeds responsible for antimicrobial and antioxidant activity have been identified using gas chromatography mass spectroscopy to find out the scientific basis for the traditional beliefs that black cumin seeds possess antimicrobial and antioxidant properties.

Table (2): Effect of common fennel concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	2.8±0.11	10.5±0.41	12.2±0.39	13.8±0.35
<i>Bacillus cereus</i> ATCC 10987	10.9±0.18	12.5±0.44	14.3±0.45	16.0±0.42
<i>Bacillus subtilis</i> ATCC6633)	0	0	0	0
<i>E.coli</i> (O157 : H7)	3.9±0.14	8.9±0.30	12.2±0.31	13.0±0.27
<i>Listeria monocytogenes</i> ATCC 51778	7.0±0.21	8.1±0.26	13.3±0.29	13.9±0.29
<i>Salmonella typhimurium</i> ATCC 2556	7.5±0.25	9.2±0.22	15.0±0.50	17.7±0.43
<i>Staph.aureus</i> ATCC 29740	9.8±0.32	11.0±0.10	12.2±0.33	13.0±0.28
<i>Yersinia enterocolitica</i> ATCC (23715)	0	0	0	0

Table (3): Effect of black-cumin concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	0	0	0	0
<i>Bacillus cereus</i> ATCC 10987	0	0	0	0
<i>Bacillus subtilis</i> ATCC6633)	6.0±0.12	8.2±0.20	9.9±0.31	11.9±0.34
<i>E.coli</i> (O157 : H7)	0	0	0	0
<i>Listeria monocytogenes</i> ATCC 51778	0	0	0	0
<i>Salmonella typhimurium</i> ATCC 2556	0	0	0	0
<i>Staph.aureus</i> ATCC 29740	8.8±0.18	9.4±0.24	12.0±0.33	13.3±0.37
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (4) shows the effect of Coriander concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). Coriander had some antibacterial effects against *Bacillus cereus*, *E. coli* (O157: H7), *Listeria monocytogenes*, and *Salmonella typhimurium*. While, Coriander concentration had no antibacterial effects on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was found against *Bacillus cereus* and *E. coli*. Generally, by increasing the Coriander concentration the antibacterial activity increased.

Table (4): Effect of Coriander concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	7.7±0.23	8.2±0.10	10.9±0.11	12.3±0.26
<i>Bacillus cereus</i> ATCC 10987	9.2±0.25	11.0±0.12	12.9±0.24	14.4±0.32
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	9.2±0.42	10.8±0.19	12.1±0.23	13.3±0.45
<i>Listeria monocytogenes</i> ATCC 51778	5.5±0.31	8.3±0.18	11.0±0.21	12.7±0.34
<i>Salmonella typhimurium</i> ATCC 2556	6.5±0.14	8.2±0.26	9.9±0.10	10.6±0.20
<i>Staph.aureus</i> ATCC 29740	0	0	0	0
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

The effect of black-pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm) in Table (5) shows that the inhibition of dairy spoilage and poisoning bacteria increased by increasing the thyme concentration. Also, all black-Pepper concentrations had no inhibitory effect on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. Generally, *Bacillus cereus* and *E. coli* (O157: H7), and were totally inhibited by the black-Pepper concentration.

The effect of rosemary concentration (% w/vol) on certain different dairy spoilage and poisoning bacteria (zone of inhibition in mm) was tabulated in Table (6). It could be observed that Rosemary concentration was found to have antibacterial activity against all of the tested dairy spoilage and poisoning bacteria, inhibition of *Yersinia enterocolitica* and the antibacterial activity increased with increasing of rosemary concentration.

Table (5): Effect of Black-Pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	9.8±0.20	11.9±0.31	14.9±0.43	15.5±0.40
<i>Bacillus cereus</i> ATCC 10987	1.8±0.10	2.2±0.15	4.3±0.17	5.9±0.17
<i>Bacillus subtilis</i> ATCC 6633	9.1±0.19	10.9±0.23	12.0±0.21	13.2±0.33
<i>E.coli</i> (O157 : H7)	10.0±0.22	13.8±0.26	16.9±0.44	17.6±0.45
<i>Listeria monocytogenes</i> ATCC 51778	7.0±0.17	9.8±0.22	11.5±0.20	13.3±0.28
<i>Salmonella typhimurium</i> ATCC 2556	9.8±0.21	12.0±0.24	14.3±0.31	14.9±0.30
<i>Staph.aureus</i> ATCC 29740	8.0±0.18	9.9±0.19	12.3±0.30	14.0±32
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (6): Effect of rosemary concentration (% w/vol) on some different spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	9.8±0.20	11.9±0.31	14.9±0.45	15.5±0.33
<i>Bacillus cereus</i> ATCC 10987	1.8±0.10	2.2±0.11	4.3±0.17	5.9±0.10
<i>Bacillus subtilis</i> ATCC 6633	9.1±0.18	10.9±0.13	12.0±0.23	13.2±0.31
<i>E.coli</i> (O157 : H7)	10.0±0.22	13.8±0.22	16.9±0.32	17.6±0.41
<i>Listeria monocytogenes</i> ATCC 51778	7.0±0.19	9.8±0.32	11.5±0.28	13.3±0.32
<i>Salmonella typhimurium</i> ATCC 2556	9.8±0.20	12.0±0.41	14.3±0.30	14.9±0.29
<i>Staph.aureus</i> ATCC 29740	8.0±0.18	9.9±0.20	12.3±0.32	14.0±0.31
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

The data in Table (7) demonstrates the effect of cayenne Pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). Cayenne pepper had some antibacterial effects against *Staph.aureus*, *Salmonella typhimurium*, *E. coli*, and *Bacillus cereus*. Whereas, cayenne Pepper concentration had no antibacterial effects on *Bacillus subtilis*, *Listeria monocytogenes*, and *Yersinia enterocolitica*, and were totally inhibited by the cayenne Pepper concentration. The aqueous and methanolic extracts from *Amomum aromaticum*, *Brassica nigra*, *Cinnamomum zeylanicum*, *Curcuma longa*, *Mentha longifolia* and *Z. officinale*; aqueous extract from *Majorana hortensis*; and methanolic extracts form *Myristica fragrans* and *Syzygium aromaticum* inhibited the growth of *Staphylococcus aureus*. Aqueous and methanolic extracts from *Coriandrum sativum*, *Curcuma longa*, *P. nigrum*, *Syzygium aromaticum*, *Trigonella foenum-graecum* and *Z. officinale*; aqueous extracts from *Myristica fragrans*, *Cuminum cyminum* and *Elettaria cardamomum*; and methanolic extracts from *Amomum aromaticum*, *B. nigra*, *Majorana hortensis* and *Trachyspermum ammi* were effective against *Aspergillus flavus*

Table (7): Effect of Cayenne pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	4.9±0.11	6.6±0.22	8.1±0.23	10.0±0.21
<i>Bacillus cereus</i> ATCC 10987	9.9±0.19	12.0±0.28	14.9±0.41	16.0±0.25
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	8.0±0.17	10.0±0.31	11.9±0.33	12.7±0.30
<i>Listeria monocytogenes</i> ATCC 51778	6.9±0.12	9.2±0.21	11.9±0.31	13.3±0.33
<i>Salmonella typhimurium</i> ATCC 2556	4.0±0.10	9.9±0.20	12.2±0.29	13.4±0.34
<i>Staph.aureus</i> ATCC 29740	5.1±0.20	7.4±0.19	8.8±0.16	10.0±0.20
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (8) reveals the effect of ginger concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). It could be noticed that the inhibition of dairy spoilage and poisoning bacteria increased by increasing the thyme concentration. Also, all thyme concentrations had no inhibitory effect on *Bacillus subtilis* and *Yersinia enterocolitica*. Generally, *Bacillus cereus*, *E. coli* (O157: H7), and *Listeria monocytogenes* were totally inhibited by the Ginger concentration.

Table (9) shows the effect of red-Pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). Red-pepper had some antibacterial effects against *Bacillus cereus*, and *Salmonella typhimurium*. Whereas, red-pepper concentration had no antibacterial effects on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was found against *Bacillus cereus* and generally, by increasing the red-Pepper concentration the antibacterial activity increased.(Parihar *et al.*, 2007).

Table (8): Effect of Ginger concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	7.0±0.23	8.4±0.12	10.0±0.20	12.3±0.25
<i>Bacillus cereus</i> ATCC 10987	9.2±0.25	11.0±0.17	12.9±0.25	14.4±0.37
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	1.7±0.11	4.8±0.10	7.7±0.17	9.6±0.21
<i>Listeria monocytogenes</i> ATCC 51778	2.2±0.15	5.6±0.15	10.0±0.19	12.4±0.31
<i>Salmonella typhimurium</i> ATCC 2556	6.5±0.18	8.7±0.18	10.7±0.20	12.0±0.30
<i>Staph.aureus</i> ATCC 29740	0	0	0	0
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (9): Effect of red-pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	7.0±0.12	8.4±0.18	10.0±0.27	12.3±0.31
<i>Bacillus cereus</i> ATCC 10987	9.2±0.11	11.0±0.21	12.9±0.29	14.4±0.39
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	1.7±0.10	4.8±0.11	7.7±0.17	9.6±0.19
<i>Listeria monocytogenes</i> ATCC 51778	2.2±0.12	5.6±0.16	10.0±0.20	12.4±0.33
<i>Salmonella typhimurium</i> ATCC 2556	6.5±0.14	8.7±0.21	10.7±0.21	12.0±0.40
<i>Staph.aureus</i> ATCC 29740	0	0	0	0
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (10) express the effect of white Pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

White pepper concentration resulted in antibacterial effects against *Bacillus cereus*, *Listeria monocytogenes*, and *E. coli*. On the contrary, White Pepper concentration found to have no antibacterial activity against *Bacillus subtilis*, *Salmonella typhimurium*., *Staph.aureus* and *Yersinia enterocolitica*.

Syamala *et al.*, (2006) evaluated the antibacterial activities of extracts of 9 plant species (*Zingiber officinale*, *Azadirachta indica*, *Piper nigrum*, *Punica granatum*, *Solanum nigrum*, *Cynodon dactylon*, *Ziziphus jujuba*, *Foeniculum vulgare* and *Portulaca oleracea*) against *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus sp.* and *Mycobacterium smegmatis* by well plate and disc diffusion methods. The *Punica granatum* peel extract effectively controlled the growth of Gram positive (*Staphylococcus aureus*, *Bacillus sp.* and *M. smegmatis*) and Gram negative (*Pseudomonas*

aeruginosa and *E. coli*) bacteria. *M. smegmatis* and *Staphylococcus aureus* were the most susceptible to most of the herbal extracts. The plant materials were more effective against Gram positive bacteria than against Gram negative bacteria.

Table (11) the effect presents that the on some different turmeric had some antibacterial effects against both *Salmonella typhimurium*, *Bacillus cereus*, *Listeria monocytogenes*, *Staph.aureus*, and *E. coli*(O157:H7). Although, Turmeric concentration had no antibacterial effects on *Bacillus subtilis* and *Yersinia enterocolitica*. The highest remarkable antibacterial effect was found against *Salmonella typhimurium*, and *Bacillus cereus* Generally, by increasing the turmeric concentration the antibacterial activity increased.

Erkmen and Ozcan (2004) investigated the bacteriostatic and bactericidal effects of five essential oils, i.e. oregano (*Origanum onites*), rosemary (*Rosmarinus officinalis*), sage (*Salvia triloba* [*S. fruticosa*]), fennel (*Foeniculum vulgare* subsp. *piperitum*) and savory (*Satureja hortensis*), on bacteria (*Escherichia coli*, *Salmonella typhimurium*, *Staphylococcus aureus*, *Bacillus subtilis*, *B. cereus*, *Yersinia enterocolitica*, *Listeria monocytogenes* and *Enterococcus faecalis*), yeasts and moulds (*Saccharomyces cerevisiae*, *Candida albicans*, *Aspergillus niger* and *Rhizopus oryzae*) using the broth microdilution method with essential oil concentrations ranging from 0.025 to 5.0% (v/v) in combination with spectrophotometry. Oregano essential oil was effective against all the tested microbial strains with bactericidal and fungicidal concentrations at 0.025%. Rosemary essential oil was bactericidal on Gram-positive and Gram-negative bacteria at 0.1 and 0.3%, respectively. Fennel essential oil concentration at 0.1 and 0.3% had bacteriostatic effect on Gram-positive bacteria and Gram-negative bacteria, respectively. The highest bactericidal concentration (1.0%) of sage essential oil was observed on *Y. enterocolitica* and lowest (0.05%) on *L. monocytogenes*. Savory essential oil was bactericidal on Gram-negative bacteria at 0.5%. Gram-positive bacteria were more sensitive to essential oils than Gram-negative. Bacterial strains were generally more sensitive to the essential oils than yeasts and moulds. The use of essential oils as antiseptics and disinfectants could be worthwhile for cleaning, disinfection and sanitation of community places and food processing plant.

Table (10): Effect of white pepper concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	5.4±0.10	6.2±0.16	8.0±0.17	9.9±0.19
<i>Bacillus cereus</i> ATCC 10987	8.8±0.11	9.3±0.20	11.3±0.22	12.9±0.33
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	7.1±0.18	9.3±0.22	10.0±0.21	12.0±0.35
<i>Listeria monocytogenes</i> ATCC 51778	7.7±0.19	10.0±0.30	12.7±0.32	13.9±0.40
<i>Salmonella typhimurium</i> ATCC 2556	0	0	0	0
<i>Staph.aureus</i> ATCC 29740	0	0	0	0
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (11): Effect of turmeric concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	7.2±0.19	9.7±0.21	11.9±0.30	13.0±0.32
<i>Bacillus cereus</i> ATCC 10987	6.0±0.13	8.8±0.22	10.2±0.33	13.2±0.33
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	5.2±0.11	8.3±0.19	9.9±0.29	11.2±0.27
<i>Listeria monocytogenes</i> ATCC 51778	7.0±0.19	9.1±0.20	10.0±0.30	12.3±0.28
<i>Salmonella typhimurium</i> ATCC 2556	7.5±0.17	9.8±0.23	11.0±0.33	13.4±0.30
<i>Staph.aureus</i> ATCC 29740	6.9±0.10	9.9±0.21	12.0±0.29	13.1±0.31
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

Table (12) demonstrates the effect of cinnamon concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm). cinnamon concentration had no antibacterial effects on *Bacillus subtilis*, *Staph.aureus* and *Yersinia enterocolitica*. While, cinnamon had some antibacterial effects against *Listeria monocytogenes*, *Bacillus cereus*, *E. coli* and *Salmonella typhimurium*. Generally, by increasing the cinnamon concentration the antibacterial activity increased.

Cinnamon was found to be the most effective spice against the tested microorganisms (*Staphylococcus aureus* ATCC 25923, *Klebsiella pneumoniae* FML 5, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 15753, *Mycobacterium smegmatis* CCM 2067, *Micrococcus luteus* A 2971, and *Candida albicans* ATCC 60192) using the disc diffusion method. While the weakest antimicrobial activity was displayed by fennel. Crushed red pepper and anise were found to be ineffective against the test strains (Agaoglu *et al.*;2007)

Sarkinas and Jasutiene (2007) found that the cassia essential oil affected the *B. subtilis* vegetative cells in the liquid medium, but did not destroy the spores of the bacteria. It affected the numbers of *Salmonella typhimurium*, but did not destroy *Salmonella enteritidis*. The cinnamon essential oil was efficient for yeast culture; it inhibited the growth of the test culture on the entire plate. The curcuma extract had a slight inhibitory effect on *L. monocytogenes*; this bacterium could grow at a low temperature. Generally, *Bacillus cereus* and *E. coli* (O157: H7), and were totally inhibited by the black-Pepper concentration. In conclusion, Rosemary and Ginger, Thyme, and Common fennel found to be the most inhibitory and could be recommended in dairy products production.

Therefore, it could be recommended that the most inhibitory effect was in Ginger, Thyme, Rosemary, and Common fennel. So, these spices and herbs could be used in the production of some dairy products with extended shelf life and high microbiological quality along refrigerated storage.

Table (12): Effect of cinnamon concentration (% w/vol) on some different dairy spoilage and poisoning bacteria (zone of inhibition in mm).

spoilage and poisoning bacteria	Zone of inhibition (mm) with different concentrations			
	0.1 %	0.2 %	0.3 %	0.4 %
<i>Bacillus cereus</i> ATCC 14049	5.8±0.10	8.0±0.21	9.1±0.28	10.0±0.20
<i>Bacillus cereus</i> ATCC 10987	1.8±0.11	5.1±0.18	7.7±0.26	9.0±0.19
<i>Bacillus subtilis</i> ATCC 6633	0	0	0	0
<i>E.coli</i> (O157 : H7)	4.3±0.18	7.0±0.14	8.2±0.22	10.0±0.28
<i>Listeria monocytogenes</i> ATCC 51778	6.0±0.19	8.9±0.19	10.0±0.27	12.7±0.30
<i>Salmonella typhimurium</i> ATCC 2556	2.7±0.14	6.1±0.20	8.8±0.25	10.9±0.28
<i>Staph. aureus</i> ATCC 29740	0	0	0	0
<i>Yersinia enterocolitica</i> ATCC 23715	0	0	0	0

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التأثير المضاد للبكتريا للأعشاب والتوابل المستخدمة في صناعة بعض منتجات الألبان

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تم دراسة التأثير المضاد للبكتريا المرضية والمسببة للفساد لاثني عشرة عشبة وتابل هي: الروزمارى ، زعتر ، زنجبيل ، شمر ، الكزبرة ، حبة البركة ، الفلفل الأسود ، فلفل أحمر ، الفلفل الأبيض ، الكركم ، والقرفة ، والشطة الحمراء بتركيز 0.1 و 0.2 و 0.3 و 0.4% ضد أكثر ثمانية ميكروبات مسببة للفساد والتسمم البكتيرى فى الألبان وهى:

<i>Bacillus cereus</i> a ATCC 10987	<i>Bacillus cereus</i> b ATCC 14049
<i>Bacillus subtilis</i> ATCC 6633	<i>E.coli</i> O157:H7
<i>Listeria monocytogenes</i> ATCC 51778	<i>Salmonella typhimurium</i> ATCC 2556
<i>Staphylococcus aureus</i> ATCC 29740	<i>Yersinia enterocolitica</i> ATCC 23715

وهذا باستخدام طريقتى *Agar well diffusion* و *disc assay* لمعرفة النشاط المضاد للبكتريا لإثني عشر عشبة خام وتابل حيث تم قياس قطر الهالة الرائقة حول العشب لمعرفة القدرة التثبيطية له. وقد خلصت النتائج الي أن :

لم يكن لأى من تركيزات الزعتر المستخدمة أى قدرة تثبيطية على كل من سلالات *Bacillus subtilis* وكذلك *Yersinia enterocolitica* بينما حدث تثبيط بالكامل لكل من سلالات *E.coli* (O157: H7), *Listeria monocytogenes* *Staph.aureus* مع كل تركيزات الزعتر المستخدمة. في حين أن كل تركيزات الشمر ليس لها أى تأثير مضاد لسلالات بكتريا *Bacillus subtilis* و *Yersinia enterocolitica*. بينما كان أعلى تأثير مضاد للبكتريا ضد *Bacillus cereus* و *Staph.aureus*، وبزيادة تركيزات الشمر إزدادت القدرة التثبيطية المضادة للبكتريا. كما كان الشمر من أفضل المواد تثبيطاً لسلالات *Bacillus cereus*. وعلى العكس من ذلك ، وجد أن تركيزات حبة البركة ليس له أى نشاط مضاد للبكتريا ضد السلالات المختبرة المسببة لفساد وسمية منتجات الألبان. وجد أن الكزبرة لها بعض الآثار المضادة للبكتيريا ضد *Listeria monocytogenes*, *Salmonella typhimurium* , وكذلك *Bacillus cereus* (O157:H7) أما أعلى تأثير ملحوظ مضاد للجراثيم فى *Bacillus cereus*

وجد أن معدل التثبيط للميكروبات إزداد بزيادة تركيز الزعتر والروزمارى والزنجبيل والشمر التى أظهرت أعلا معدلات تثبيط للبكتريا المرضية والمسببة للفساد. بينما جميع تركيزات الفلفل الأسود لم يكن له تأثير تثبيطى على كلا من *Bacillus cereus*، *Staph.aureus* وكذلك *Yersinia enterocolitica*.

لهذا يمكن التوصية باستخدام أى من الروزمارى والزنجبيل والزعتر أو الشمر كمواد تساعد على زيادة قوة الحفظ فى منتجات الألبان بالإضافة إلى إكسابها طعماً وألوان جديدة مرغوبة ومقامة تواجد ميكروبات مرضية .