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Annals of Biological Research, 2011, 2 (6):22-30
(http://scholarsresearchlibrary.com/archive.html)

Annals of Biological Research
Scholars Research Library

ISSN 0976-1233
CODEN (USA): ABRNBW

Extend the Shelf-life of Different Natural Packed Juices by Electromagnetic Induction

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ABSTARCT

The effect of high frequency electromagnetic field has been used for inactivation pathogen microorganism of different packed natural juices(juice of apple , juice of peach , juice of apricot).All samples of juices were filled in pouches then electromagnetic induction which discharges square-wave pulses with variable voltage 1-20 kV/cm and different frequency(2-3.5GHz , 3.5-5 GHz , 5-6.5GHz) have been used. Pure cultures of E. coli O160: H6 (Yerevan, Armenia), Salmonella Enteritidis 1.23 (Yerevan ;Armenia) L. monocytogenes 1.91 (Yerevan ;Armenia) were used to inoculate (1ml) with samples before EMI to scientifically prove the usage of electromagnetic field for sterilization of these packed fruit juices against population of pathogen microorganism, so we want to experimentally show that EMI can substitute thermal processing in conservation industries. According to" Wald test" the effect of p-value for type of juices , type of treatments ,and for inactivation E-coli ,Salmonella Enteritidis and L. monocytogenes have significant level (p-value=0,001) . Microbiological shelf-life were also determined in these natural fruit juices during 2 month which have been stored at 5 ^{o}c in order to ensure about the safety of pouches.

Key words: electromagnetic induction (EMI), flexible packaging, juice of peach, juice of apple, juice of apricot, pathogen microorganism

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INTRODUCTION

Consumption of natural juices can provide health benefits due to the antioxidant and high capacities of their vitamins and minerals [2,25]. However, these products without an efficient processing may be potential source of microbiological diseases, the low to middle acidity and high

water activity (0.85–0.90) of these fruits can favor the growth of pathogenic microorganisms. [19,21,29]. Pathogenic microorganisms such as Salmonella spp.,Escherichia coli and Listeria monocytogenes can be transferred to the edible part and juice causing diseases [30,28], which survival and growth of these bacteria have been demonstrated by several researchers [6, 26]. Although, thermal treatment effectively destroys pathogenic microorganisms in fruit juices, undesirable changes on the organoleptic and nutritional properties of juices are observed [3, 4,20]. For that reason, significant efforts are leading to the development of novel on-thermal processes such high frequency electric field, an alternative preservation process that improving to be able to inactivate spoilage and pathogenic microorganisms without significantly affect the organoleptic and nutritional properties of several foods [14, 15,18, 20]. The usage of high frequency (2-15GHz) with different voltage (1-20kV/cm)(18,23) for fluid foods placed between two electrodes in batch flow systems using low processing temperatures (near 40 ^{o}c) and low energy efficiency for sterilization with regard to the thermal treatment [20,23,25]. This frequency allocated by federal communication commission (FCC)(16, 17, 23,27).The effectiveness of EMI treatment to reduce Salmonellaspp., E. coli and L. monocytogenes population in some fruit juices has been reported later [5, 22]. The microbiological shelf-life extension of some PEF-treated acid juices such as orange, cranberry and tomato in comparison with unprocessed juices (control) has also been demonstrated [3,5,8]. However, the effect of EMI treatment on the naturally occurring microorganisms in packed juices such as these fruit has not been found in the literature.

The objectives of this study are scientifically proved and experimentally shown the usage of electromagnetic field for sterilization of these packed fruit juices against populations of E. coli, Salmonella Enteritidis and L. monocytogenes inoculated in, as Zhang says, “Our work has improved food safety by enabling the food industry to make better decisions about how to reduce or eliminate pathogens microorganism [7]. We want to prove EMI can substitute thermal processing in conservation industries, and have a lot of privilege [33-36].

MATERIALS AND METHODS

2.1. Preparation of fruit juices

Peach and apricot and apple fruits at commercial ripeness were chosen from a supermarket of Tehran -Iran. The fruits were washed, dried, cut into slices and made juice through TEFAL blender (Model 8110 BSA,Brazil). Peach and apricot and apple juices were centrifuged at 12,500 rpm for 15 min at 4 ^{0}C in an Avanti TM J-25 Centrifuge (Beckman Instrument, Inc,USA). The supernatant juice was filtered .Three kind of samples were prepared with pure cultures of E. coli, Salmonella Enteritidis , L. monocytogenes inoculated (1ml) in before EMI: [33, 34]

1-Pouches contain 100 ml juice of apple
2-Pouches contain 100 ml juice of peach
3-Pouches contain 100 ml juice of apricot

All pouches were filled with samples of natural fruit, after sealing of pouches, different condition of EMI (2-3.5GHz , 3.5-5 GHz , 5-6.5GHz) have been done for them. Analytical parameters such as pH (Crison 2001 pH meter; Crison Instruments, SA, Barcelona, Spain) soluble solid

content (Atago RX-1000 refract meter; Atago Company Ltd., Japan), sealer (Impulse Sealer, Manual Instruction ,Korea) were measured according to the ISIRI Regulation [33-36]

2.2. Microbial culture

Pure cultures of E. coli O160: H6 (Orbeli Institute of Physiology, Yerevan, Armenia), Salmonella Enteritidis 1.23 (National Institute of standard CJSC,Yerevan ;Armenia) L. monocytogenes 1.91 (National Institute of standard CJSC,Yerevan ;Armenia) were used for this study. Strains of E. coli and L. monocytogenes were grown in type tone soy broth (TSB) (Biokar Diagnostics;Beauvais, France) plus 0.6% (w/v) of yeast extract (Biokar Diagnostics); whereas, strain of S. Enteritidis was cultured in TSB. E. coli and Salmonella Enteritidis and L.monocytogenes were incubated at 37 ^{o}C with continuous agitation at 120 rpm for15 h, . The final concentration achieved of microorganisms in the growth media was approximately109–1010 colony forming units/ml (CFU/ml).Each samples of peach, apricot and apple juices (100 ml) to be treated with or without EMI was inoculated with 1ml of each pure culture of pathogenic microorganism to obtain thus, the final concentration of 107–108 CFU/ml approximately in each sample of juices. The growth of bacteria in samples have been showed as response (non parametric: negative or positive) [33-36]

2.3. High frequency electromagnetic field and processing parameters

A continuous flow high frequency electromagnetic model pilot-scale [1,11-13] which discharges with square-wave pulses was used for sterilization of these packed fruit juices [11]. Inner part of system composed electromagnetic induction, water bath, and stainless-steel tube submerged in water bath, variable pump .Electromagnetic induction containing: A:capacitor: balance of voltage ; B: fuse: safety of system; C:diode: safety of system; D:magnetron: source of frequency's; F:transformation: change of voltage 1-20kV/cm [9,10] with different frequency(2-15 GHz). The packed juices were put in treatment chamber with volume 60 lit (W=40cm,L =60cm,H=25 cm).

The full intelligent PLC composed 30 memories to chose different programming of voltage and frequency pulse. Total usage of power (7-21 KW) was controlled through of a Pulse generator, which the excessive decrease of usage energy in comparison with other system; the flow rate (300-400 ml/sec) was adjusted by gear pump. Other technological specification is complete isolation system of environment, two intelligent micro processor for controlling electromagnetic induction and critical point of system so The temperature during electromagnetic induction did not exceed 40 ^{o}C, as you see in fig 1[33-36].

The applied residence time in this chamber was calculated according to Yang et al. [31] as follows:

$$\mathbf{T_R = V_C / F_r}$$

V_c is the volume of a chamber (cm^3) and F_r flow rate (ml/s)which estimate 3-5 min (20 sec induction, 20 sec rest) 2 pulse per min

A **B**

Fig. 1 (A)-Electromagnetic field, (B) -Panel control, [1]

2. 4-Samples packaging and storage

Unprocessed and processed of different packed fruit juices were filled (leaving the minimum amount of headspace volume) and packaged in one type of multilayer flexible pouch [33-36]. The properties of this container (3 Layers) were shown in table 1

Table 1- Analytical characteristics of container (multilayer flexible pouch) [24]

Sample	Layers	Tensile of film	Tensile of sealing film	O.T.R(ml/m ^{2}.day) Oxygen transmit ion rate	W.V.T.R(g/m ^{2}.day) Water transmit ion rate
PET\AL\LLD	12\12\100	65.16	*46.403*	*0*	*0.123*

***PET**: Poly Ethylene Terphetalat; **LLD**: Low Density Poly Ethylene; **Al**: Aluminum*

Statistical Analysis

Multilevel factorial design was carried out in these samples which inoculated in different condition with EMI or without EMI ,so we must find a model for relationship between type of juices, type of treatment , and type of microorganism .We have described this variables (microorganisms) with frequency tables; cross tables and relative diagrams ,and for deduction of these variables have used "logistic regression" and "add ratio"; as a large amount of positive number of microorganisms in EMI treatment suspected positive growth of microorganisms in media culture evaluate negative ,in order to obtain model of logistic regression. The effect of each period of frequency in electromagnetic field have been done , which was showed in table 2 [33-36], so we have for juice of apple 4 treatments which were renamed A1 –A4, and for juice of peach 4 treatments which were renamed B1-B4, , and for juice of apricot 4 treatments which were renamed D1-D4 .Each treatment was repeated in 3 run [33-36],as you see in table 2,fig 2, fig 3,fig 4

RESULTS

In this study, electromagnetic field with variable voltage 1-20 kV/cm and frequencies (2-3.5GHz, 3.5-5 GHz , 5-6.5GHz) was used according to previous researches [1,11-13, 32-36]. The best result for inactivation of microorganism belong to 5-6.5 GHz. [1, 11-13].

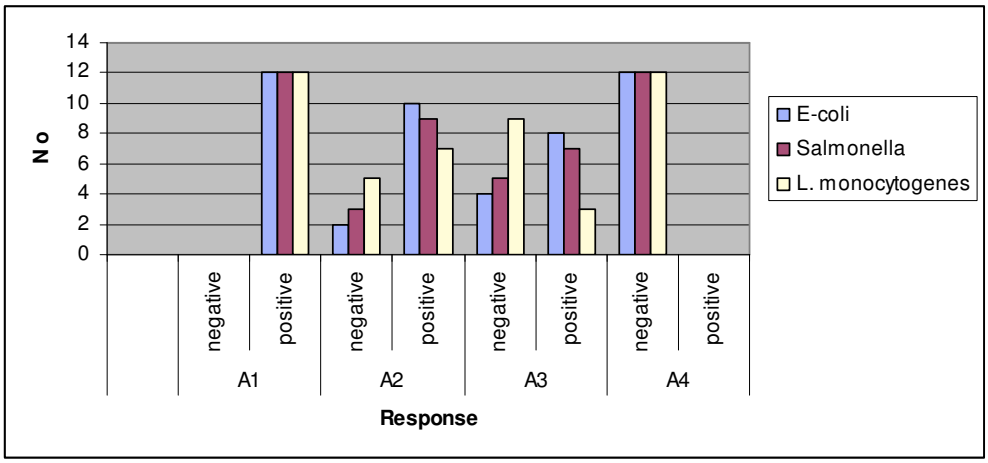

Fig 2 .Effect of EMI for inactivation of *E-coli* ,*Salmonella* and L. monocytogenes in apple juice

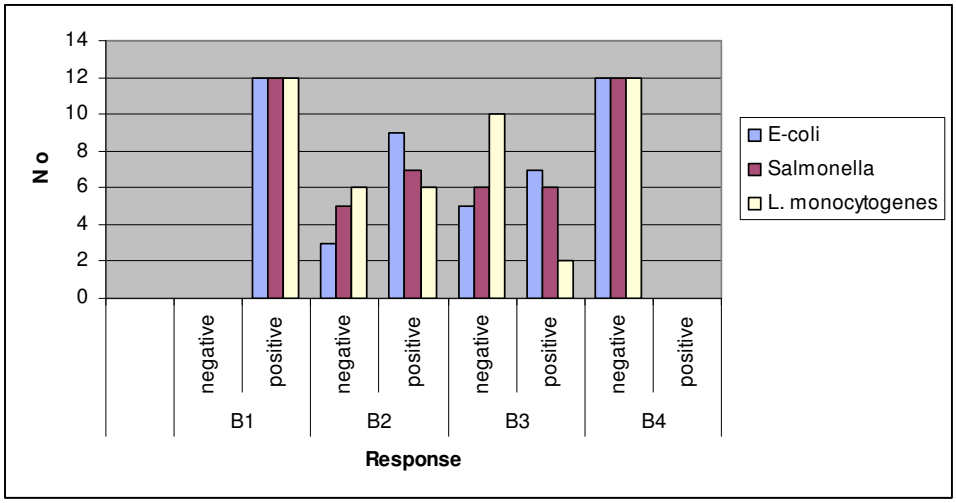

Fig 3. Effect of EMI for inactivation of *E-coli* , *Salmonella* and L. monocytogenes in peach juice

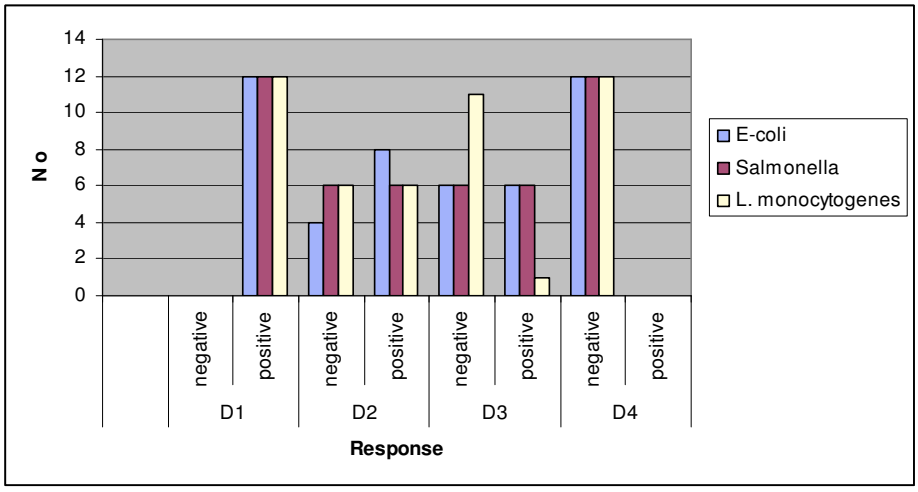

Fig 4 . Effect of EMI for inactivation of *E-coli, Salmonella* and L. monocytogenes in apricot juice

Table 2-The Effect of EMI for inactivation of pathogen microorganisms in different packed natural fruit juices

Treatment	Response	*E. coli*	*Salmonella Enteritidis*	L. monocytogenes	Treatment renamed
Apple juice(control)	negative	0	0	0	A1
	positive	12	12	12	
Apple juice +(2-3.5 GHz) EMI	negative	2	3	5	A2
	positive	10	9	7	
Apple juice +(3.5-5 GHz) EMI	negative	4	5	9	A3
	positive	8	7	3	
Apple juice +(5-6.5 GHz) EMI	negative	12	12	12	A4
	positive	0	0	0	
Peach juices +(control)	negative	0	0	0	B1
	positive	12	12	12	
Peach juices +(2-3.5 GHz) EMI	negative	3	5	6	B2
	positive	9	7	6	
Peach juices +(3.5-5 GHz) EMI	negative	5	6	10	B3
	Positive	7	6	2	
Peach juices +(5-6.5 GHz) EMI	negative	12	12	12	B4
	positive	0	0	0	
Apricot juices +(control)	negative	0	0	0	D1
	positive	12	12	12	
Apricot juices +(2-3.5 GHz) EMI	negative	4	6	6	D2
	positive	8	6	6	
Apricot juices +(3.5-5 GHz) EMI	negative	6	6	11	D3
	positive	6	6	1	
Apricot juices +(5-6.5 GHz) EMI	negative	12	12	12	D4
	positive	0	0	0	

Shelf Life

All pouches finally were stored in refrigerator at 5 ^{o}c in order to determine the shelf life of different natural packed fruit juices which have been inoculated with pure cultures of E. coli, Salmonella Enteritidis ,and L. monocytogenes in this container. The best storage time for each pouches were reported (after EMI sterilization), as you see in table 3 [34]

Table 3- The estimated shelf life for different packed fruit juices in this container [24]

Sample	Layers	Thickness, (µ)	Storage Time(Month) Apple juice	Storage Time(Month) Peach juice	Storage Time(Month) Apricot juice
PET\AL\LLD	12\12\100	124	2	2	2

CONCLUSION

We have obtained these results with" logistic regression" and "add ratio" which were shown in table 4 ;table 5;table 6

Table 4-Effect of high frequency electromagnetic induction for inactivation E-coli

Condition	Coefficient	Degree of freedom	P-value (Sig)	(Chance) add ratio
constant	-6.33	1	0.00	-
Type of juices	2.38	1	0.00	93.44
Type of treatment	0.73	1	0.00	0. 442

Model of logistic regression is written

Logit (be negative) = -6.33 + 2.38(type of juice) +0.73 (type of treatment)

According to" Wald test, effect of p-value for type of juices, type of treatment has significant level (p-value=0,001) for inactivation E-coli ,as you see these chances in table 4[33-36]

Table 5-Effect of high frequency electromagnetic induction for inactivation Salmonella Enteritidis

Condition	Coefficient	Degree of freedom	P-value (Sig)	(Chance) add ratio
constant	-6.24	1	0.00	-
Type of juices	4.95	1	0.00	101.87
Type of treatment	0.81	1	0.00	0. 493

Model of logistic regression is written

Logit (be negative) = -6.24 + 4.95(type of juice) +0.81(type of treatment)

According to" Wald test', effect of p-value for type of juices, and type of treatment has significant level (p-value=0.001) for inactivation Salmonella Enteritidis ,as you see these chances in table 5 [33-36]

Table 6-Effect of high frequency electromagnetic induction for inactivation L. monocytogenes

Condition	Coefficient	Degree of freedom	P-value (Sig)	(Chance) add ratio
constant	-6.72	1	0.00	-
Type of juices	5.11	1	0.00	116.07
Type of treatment	0.85	1	0.00	0.505

Model of logistic regression is written

Logit (be negative) = -6.72 + 5.11(type of juice) +0.85(type of treatment)

According to" Wald test', effect of p-value for type of juices, and type of treatment has significant level (p-value=0.001) for inactivation L. monocytogenes ,as you see these chances in table 6 [33-36]

Acknowledgements
The author would appreciate ASk Com (Afra Sanat Kimia Engineering Com, Mashad-Iran), PMA Com(Plastic Mashine Alvan, Tehran-Iran) for supplying material, and Dr. Hossien Khabazi (Afra Sanat Kimia Engineering Com, Mashad-Iran)

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