

## **Antibacterial Effects of Lactic Acid Bacteria Cell Free Extract on the Growth of Local Isolates of *Listeria monocytogenes* from some Food Sources**

**Zainab M. Alzubaidy\***

*Department of Food Technology*

*College of Agriculture*

*University of Salahaddin*

**Shean I. Kakey**

*Department of Biology*

*Faculty of Science*

*University of Koya*

**Jassim F. Ali**

\*Email: [zalzubaidii@yahoo.com](mailto:zalzubaidii@yahoo.com)

(Received 13 / 3 / 2013 ; Accepted 26 / 6 / 2013)

### **ABSTRACT**

The present study aimed to determine the antibacterial activity of lactic acid bacteria metabolites against *Listeria monocytogenes* which was isolated from some foods in local markets of Erbil and Koya cities; 225 food samples were examined for the presence of *Listeria* species particularly *L. monocytogenes* from October 2010 to March 2011. The studied samples included 70 samples of raw chicken meat, 55 samples of raw milk, 50 samples of cheese and 50 samples of raw red meat. According to motility test, hemolysin production, sugar fermentation test, 8 isolates of *L. monocytogenes* were identified, 1 (2 %) isolate from cheese, 2 (4 %) isolates from red meat and 5 (7.3 %) isolates from chicken meat. The percentages of contaminated foods for *Listeria* species from chicken meat, raw milk, cheese and red meat were 20, 9, 20 and 22% respectively. With respect to the biocontrolling attempt, the effect of cell free extract (CFE) of lactic acid bacteria (LBA) which is considered as safe against isolated *L. monocytogene* was used as biocontrolling agent. The CFE of lactic acid bacteria which isolated from local dairy products, was tested for its controlling ability by agar diffusion method, the CFE showed inhibition zone from (15-23 mm) in diameter. Also the minimum inhibitory concentration MIC of CFE was determined at three additive percentages (0.5, 1.0 and 1.5ml to 10 ml of nutrient broth), the inhibition percentage ranged from (28.75 - 48.97) at (0.5 %), (52.13-95.55) at (1 %) and (82.93 -98.15) at (1.5 %) additive percentage of CFE of LAB.

**Keywords:** *Listeria monocytogenes*, Lactic Acid Bacteria



temperatures. Although it commonly exists in raw foods of both plant and animal origin, it is also present in cooked foods due to post-processing contamination, from food processing environments; especially those that are cool and wet (Tompkin, 2002). There are seven species within the genus *Listeria* (*L. monocytogenes*, *L. ivanovii*, *L. innocua*, *L. seeligeri*, *L. welshimeri*, *L. grayii*, and *L. murrayi*), only two, *L. monocytogenes* and *L. ivanovii*, are pathogenic, causing disease in both humans and animals (Roberts and Greenwood, 2003; Swamina, 2006). Almost 99% of human listeriosis has resulted from the consumption of contaminated foods (Mead *et al.*, 1999). Listeriosis is among the most common bacterial food-borne pathogens worldwide and the disease emerging as an important food-borne pathogen of public health concern (Chukwu *et al.*, 2007) *L. monocytogenes* causes listeriosis which can be a non-invasive disease, but primarily occurs in an invasive form. It affects those with a severe underlying disease or condition, as immunosuppression and HIV/AIDS, pregnant women, unborn or newly delivered infants, and the elderly. The clinical signs of the invasive form are flu-like illness including fever, headache, nausea, vomiting, and diarrhea appear about 12 hours or more after ingestion. After several days, the more serious symptoms appear, including meningitis, encephalitis, septicemia, and intrauterine/cervical infections that may result in spontaneous abortion in pregnant women (Swamina and Gerner, 2007; Nayak *et al.*, 2010). The term “biocontrol” refers to the use of microorganisms or their metabolites to inhibit or inactivate undesirable microorganisms in food. Biocontrol was first associated with the use of starter cultures, but metabolites and fermentation products also contribute to the safety and microbiological quality of food. Biocontrol of *L. monocytogenes* in food is achieved by adding bacteriocin-producing microorganism, bacteriocin-containing fermentate, bacteriocin crude extract, or purified bacteriocin, in fact lactic fermentations are believed to be the oldest preservation known to human kind (Ross *et al.*, 2000). Such an application i.e., the antimicrobial properties of lactic acid bacteria due to their ability to produce substance as a natural competitive means to overcome other microorganisms sharing the same ecological niche (Pattnaik *et al.*, 2005) Bacteria that produce antilisterial bacteriocins include strains of lactic acid bacteria, lactic acid bacteria produce various types of compounds such as lactic acid, acetic acid, formic acid, phenyllactic acid, diacetyl, acetoin, reuterin, reutericyclin, cyclic dipeptides, bacteriocins and other inhibitory proteinaceous compounds, 3-hydroxy fatty acids and hydrogen peroxide (Schnurer and Magnusson, 2005; Francoise, 2010).

The aims of this present study were: (1) to evaluate food in local market of Erbil and Koya cities for the presence of *Listeria* especially *L. monocytogenes* (2) to test the antimicrobial activity of LAB isolated from “local dairy products” against *L. monocytogenes*.

## MATERIALS AND METHODS

Two hundred and twenty five (225) Samples of fresh white cheese and raw milk and beef meat and poultry were collected from and around different market areas in Erbil City and Koya city for a period of 4 months and examined for the presence of *L. monocytogenes* and other species. (a) Raw milk (55 samples), (b) soft cheese (50 samples), (c) red meat (50 samples), (d) poultry meat (70 samples) were collected under sterile conditions during 2010–2011. The samples were examined in accordance with EN ISO 112 90-1 and they were repeatedly propagated in the Fraser broth, cultured on selective media PALCAM and Oxford.

Diagnosis: Colonies suspected to be *Listeria* were characterized using Gram stain; catalase reaction; umbrella-shaped motility pattern by using motility test medium; haemolysis on sheep blood agar; fermentation of mannitol, rhamnose and xylose and CAMP test performed according to Bergey's Manual of Systematic Bacteriology (Seeliger and Jones, 1987).

#### **Determination of Antibacterial Activity of the Cell Free Extract ( CFE) of LAB:**

Cell free extract of all LAB which used in this study was prepared according to (Alsheekh 1999; Hameed, 2004) as follow: Lactic acid bacteria (lactic acid bacteria (LAB) include (3) strains of *Lactobacillus lactis* (LB), (2) strains of *Pediococcus petosaceus* (PC) and (3) strains of *Lactococcus lactis* (LC) was inoculated in MRA (De Man Rogosa Sharp) broth as 2 % of broth volume and incubated in anaerobic condition at 37°C for 72h. The culture then centrifuge at 5000 rpm for 30 min., The Supernatants were sterilized by filtration through 0.22 mm membranes (Millipore).The antibacterial activity of CFE of LAB was determined by agar well diffusion method as described by (Gupta *et al.*,1998; Alsheekh,1999), 0.1 ml of approximately  $10^8$  which corresponded to a 0.5 McFarland turbidity standard solution of *L.monocytogenes* was transferred into nutrient agar and spread by L- shape spreader after one hour then well (5mm) in diameter and (5mm) in height was made by using sterilized cork porrer in nutrient agar then 0.5 ml of CFE was added into the wells, the plates were incubated at 37°C for 18h,after then the inhibition zone was measured and compared with control which contains MRS media only.

#### **Determination of Minimum Inhibitory Concentration of CFE of LAB**

The MIC of CFE of all LAB was determined, the (2) isolates of *L. monocytogenes* (1%) of  $10^5$  cfu/ml of fresh culture was inoculated in 10 ml nutrient broth containing (0.5, 1, 1.5) % CFE and incubated at 37°C for 24 h, after that serial dilution for every treatment was done, then the inhibiting activity of CFE was determined by colony count by plating (0.1 ml) for each treatment into sterilized petri dishes then TSA-YE(Liofilchem) was poured into the plates and incubated at 35°C for 24 hr., after incubation, results were recorded and the percentage of the death were determined comparing with the control treatment ( 0% of CFE), (Suskovic *et al.*, 1997).

### **RESULTS AND DISCUSSION**

Pre-enrichment for samples in half fraser broth (primary *Listeria* enrichment broth LEB1). This step helps the recovery of stressed *Listeria* cell in the food samples (Mansilla, 2008), secondary *Listeria* enrichment broth LEB11, in this broth the concentration of selective agents are double. Positive fraser broth has darkened and may be black, dark brown or dark green appeared in (Fig. 1). A negative MFB has the straw color of newly made broth, these broth contain selective supplements which inhibit unwanted bacteria in the food samples which usually contain other bacteria as well. On the agar PALCAM (PLA) plates, *Listeria* colonies appeared grey-green. The colonies had black-sunken centers with a black halo against a cherry red background as shown in (Fig. 2). *Listeria monocytogenes* breaks down the esculin in the medium to glucose and esculentin,

which forms an olive-green to black complex with ferric ions which stains the colonies of *Listeria monocytogenes*. (Walter, 2000). On Oxford agar *Listeria* showed the colonies grey–green. The colonies had black–sunken centers with that were surrounded by black halos. Selectivity is provided by the presence of lithium chloride in the formula also it is increased by adding various antimicrobial agents to the base. (Bille *et al.*, 1999; Chukwu, 2007).

### Confirmation Tests for identification of *Listeria* species Isolates

Gram stain: *Listeria* species cells appeared gram-positive rods, arranged singly, or in short chains, or in pairs at V-form angles and in groups that were parallel to each other. Other conformation include catalase, motility test, *Listeria* species gave a typical umbrella growth pattern on the motility test medium near the microaerophilic subsurface of the medium at (25C°) as shown in (Fig. 3) (Brugere-Picoux, 2008)

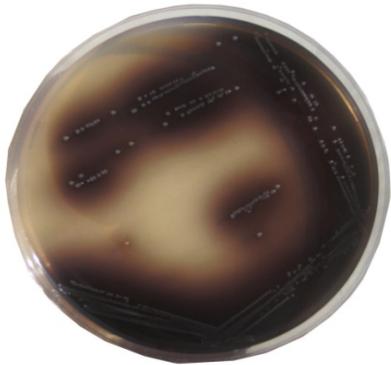


Fig. 1: *L. spp.* on Oxford agar



Fig. 2: *L. spp.* on PALCAM agar

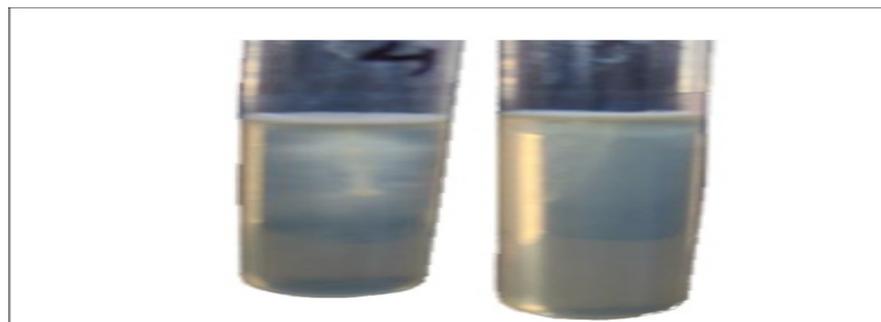


Fig. 3: Umbrella motility for *Listeria spp.*

### Biochemical examination of *Listeria monocytogenes*

All isolated *Listeria* species strains are subjected to biochemical tests for identification and to show their biochemical characteristics. The identification was confirmed by sugar fermentation. The result showed that *L.monocytogenes* fermented salcin, lactose, galactose and rhamnose, and not fermented manitol,

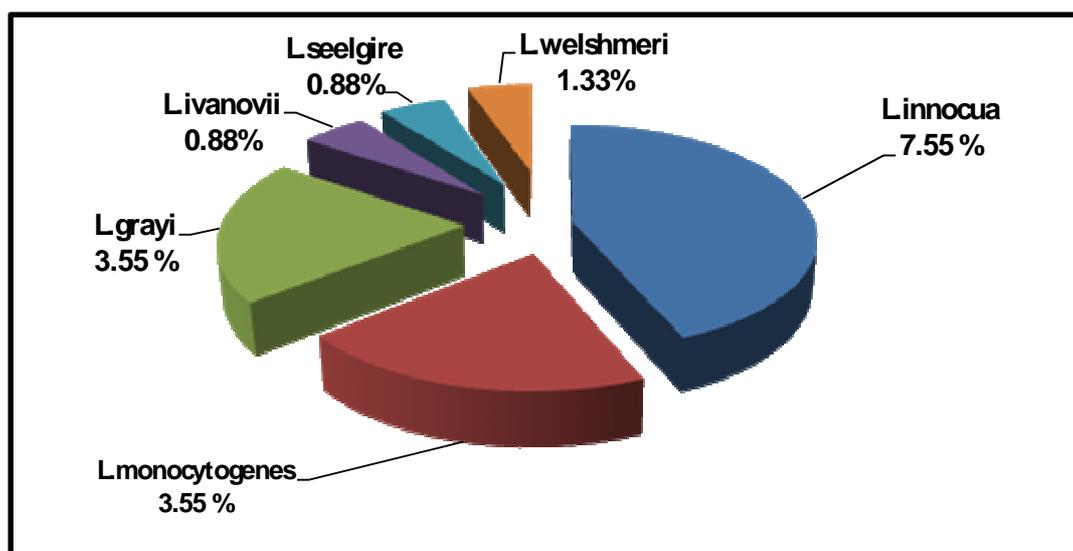
xylose, sucrose, and melbiose. All isolated strains hydrolyzed esculin to asculin, which reacts with ferric ammonium citrate producing a black precipitate coloring the media and this result agrees with (Fraser and Sperber, 1988). All isolated *L. monocytogenes* were positive for catalase and negative for oxidase.

### The presence of *Listeria* species in food samples

Bacteriological investigation was done on (225) samples of (raw poultry meat, raw milk, cheese and raw red meat). Eight isolates of *L.monocytogenes* were isolated from poultry meat, red meat and cheese samples. The incidence of *Listeria* spp. was (16.58), (39) isolates of different species of *Listeria* (*L.monocytogenes*, *L.innocua*, *L.grayi*, *L.welshmeri*, *L.ivanovii*, *L.seeligeri*), with a percentage of 3.4%, 7.23%, 3.4%, 0.85%, 0.85% and 0.85% respectively. as shown in Table (1) and (Fig. 4).

**Table 1: Incidence of *L. monocytogenes* in food samples with present occurrence in *Listeria* species**

Species	Total	Percentage
<i>L.monocytogenes</i>	8	3.4
<i>L.innocua</i>	17	7.23
<i>L.grayi</i>	8	3.4
<i>L.welshmeri</i>	2	0.85
<i>L.ivanovii</i>	2	0.85
<i>L.seeligeri</i>	2	0.85
<b>Total</b>	<b>39</b>	<b>16.58</b>



**Fig. 4: Pie chart showing the percentage of *Listeria* species in 225 food samples**

Sammarco and coworkers (2004) in Italy reported that *L. monocytogenes* was not isolated from a total of (40) raw milk samples. The incidence of *L.monocytogenes* was reported 5% in 80 raw milk samples from Ankara (Aygum and Pehlivanlar, 2006).

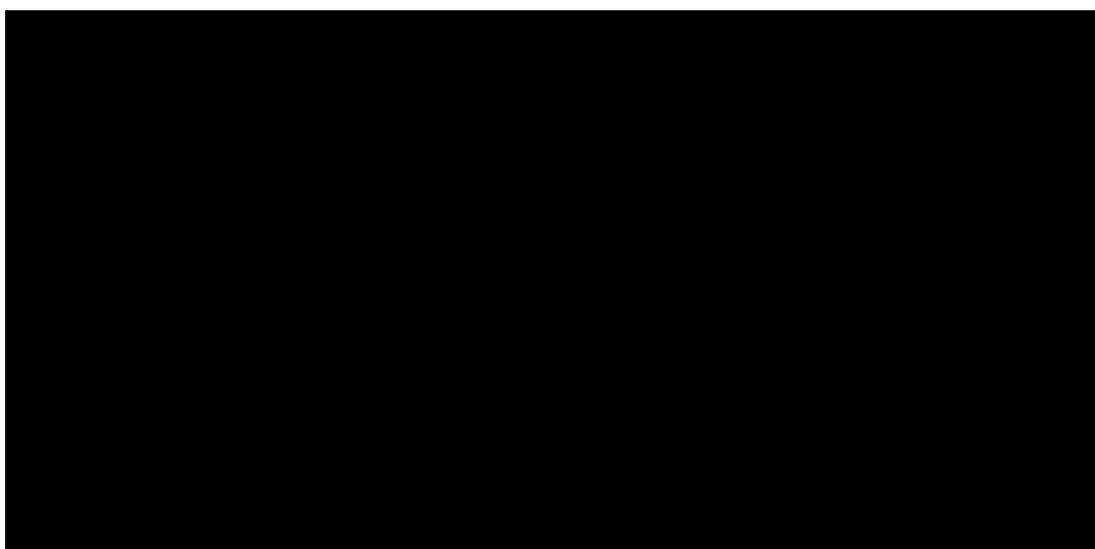
The higher prevalence of *Listeria* spp. (57.79-60.40%) and *L. monocytogenes* (49.25%) were reported by Kumar (2011) in U.S.A. Jami, S. and coworkers (2010) reported that the contamination of raw milk with *L. monocytogenes* was determined to be 4% of bulk tank milk in Iran. The occurrence of *listeria species* in (50) samples of different kinds of local cheese was 10 (20%) and *listeria monocytogenes* was 1(2%) as shown in Table (3), also *listeria innocua* was isolated from (5) samples, *L. grayi* isolated from (2) samples, *L.welshimeri* isolated from (1) sample, *L.ivanovii* isolated from (1) sample and *L.monocytogenes* was isolated from (1) sample. The incidence of *L. monocytogenes* in soft and semi-soft cheese varied from 0.50% to 46.00% (Pintado *et al.*, 2005; Colak *et al.*, 2007; Tolga *et al.*, 2010; Jakobsen *et al.*, 2011).

*L.monocytogenes* has been shown to adhere to several different food contact materials such as stainless steel, polypropylene and glass and the adhered cells show an increased resistance to cleaning agents, disinfectants and heat, all of which are used in the sanitation of the food processing plants (Dimic, 2010), therefore the contamination of cheese during processing from environment is possible. The prevalence of the organism in cheese is hazardous to consumers because cheese is ready to eat.

From a total of (70) of raw Chicken (fresh and frozen) samples 19(27.1 %) were positive for *Listeria* spp. Subdivided as 5 (7.1 %) *L.monocytogenes*, 5(7.1 %), *L.innocua*, *L. grayi* 2(2.8 %), *L.welshimeri* 1(1.4 %), 1(1.4 %) *L.seeligeri*. This result is in agreement with Ennaji *et al.*, (2008) who found that (20.3%) of poultry meat were positive for *listeria spp.*, Among the strains of *L. species* isolates, (1.3 %) *L.monocytogenes*, (16.2 %) *L. innocua*, and *L. welshimeri* (2.7 %) were identified. Abd El-Malek *et al.*, (2010) in Assuit (Egypt) found that *listeria spp.* was indicated in (52 %) and (8 %) *L.monocytogenes* in chicken legs and (54 %) and (0 %) *L.monocytogenes* in chicken fillet samples. The presence of *Listeria spp.* in (50) samples of red meat was 11(22 %), subdivided as 5(10 %) *L. innocua*, 2(4 %) *L.monocytogenes*, 2(4 %) *L.grayi*, 1(2 %) *L.welshmere*, and 1(2 %) *L.seeligeri* as shown in Table (2) and (Fig. 5). The incidence rate of *Listeria spp.* in red meat was (32 %) isolated from the examined samples. *L. monocytogenes* occurred in (4%) and *L. innocua* in (28%) of tested samples in Issuit city in Egypt (Abd El-Malek *et al.*, 2010), Nayak *et al.*, (2010) found that 10 (6.7 %) samples were positive for *Listeria species*, of which four (2.7 %) were positive for *L monocytogenes*, two (1.3 %) for *L.innocua*, three (2.0 %) for *L. seeligeri* and one (0.7 %) for *L. welshimeri*. Contamination of the meat with *L.monocytogenes* generally occurs after the slaughter and may come from the skin of the animals, the hands of the workers, the equipment and the tools used (Marinsek and Grebenc, 2002).

**Table 2: Distribution of *Listeria* spp. isolated from 225 food samples**

Type of sample	No. of sample examined	No. of positive samples for <i>Listeria</i> spp.	No. of samples for <i>L.monocytogenes</i>
Poultry meat	70	19	5
Raw Milk	55	5	0
Cheese	50	10	1
Red meat	50	11	2
<b>Total</b>	225	45	8

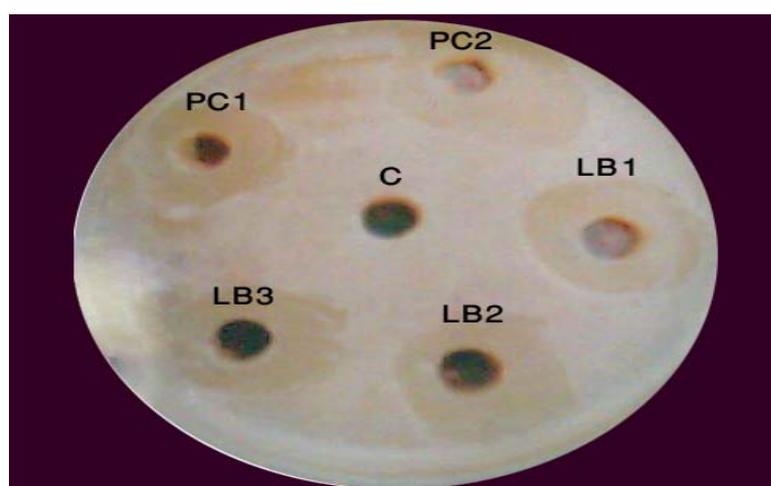
**Fig. 5: Pie chart showing incidence of *Listeria* spp. in (225) samples of chicken meat, raw milk, red meat and cheese.*****In Vitro* Antagonistic activity of Lactic Acid Bacteria against *L. monocytogenes***

Cell free extract (CFE) metabolites of lactic acid bacteria (LAB) grown in MRS media have an inhibitory effect against (2) strains of *L.monocytogenes*. By using well diffusion method, the inhibitory effect of LAB cell free extract was variable and the inhibition zone was ranged from (15-23) mm. as shown in Table (3) and (Fig. 6), and the effect of LC and LB on the growth of *L. monocytogenes* was higher than PC as shown in Table (3). The minimum inhibitory concentration of CFE of LAB against *L. monocytogenes* at (0.5,1 and 1.5) v/v of CFE which concentrated for two folds was ranged from (28 – 98 %) as shown in Table (4) the inhibition percentage increases as the concentration of CFE increases. LAB produces many antimicrobial substances like organic acids, hydrogen peroxide and bacteriocins that inhibit other bacteria and fungi (Mansilla, 2008). The products of LAB metabolites contribute, not only for preservation, but also to the flavor, aroma and texture, thereby helping to determine unique product characteristics (Pereira, *et al.*, 2008). LAB are used as natural or selected starters in food fermentations, especially for the manufacture of dairy products with functional and probiotic properties. The effect of isolates LAB against *L.monocytogenes* is very important in the dairy industry, as this pathogen was isolated from cheeses manufactured from raw milk, ripened cheeses, and in

the wash water employed for cheese production (Glass and Doyle, 2005). Svirakova *et al.*, (2009) reported that *Lactococcus lactis* caused reduction in listeria growth from the level of  $10^3$  CFU/ml to  $10^0$  CFU/ml. Nespolo and Brandelli, (2010), reported that *L. plantarum* showed antimicrobial activity against *Listeria monocytogenes*. When *Pediococcus acidilactici*, was added to frankfurters and cooked ham inoculated with *L. monocytogenes* showed bacteriostatic activity in cooked ham and bactericidal activity in frankfurters (Kumar, 2011). The use of lactic acid bacteria in fermented sausages as protective culture has a bacteriostatic or bactericidal action on *L. monocytogenes* growth. Biopreservation is an additional factor in the production of safer food but it alone cannot warrant microbiological safety (Zdolec *et al.*, 2007). Lactic acid bacteria metabolites with activity against *L. monocytogenes*, have attracted great interest in the past ten years in food science area. Particularly, bacteriocins from lactic acid bacteria (LAB) are very important because they constitute a group of industrial microorganisms that may improve sensorial properties, shelf life and safety of foods (Martinez and Martinis, 2006).

**Table 3: The inhibitory effect of CFE of LAB on growth of *L. monocytogenes* strains by well diffusion method**

LAB strains	Inhibition zone (mm)	
	<i>L.monocytogenes1</i>	<i>L.monocytogenes 2</i>
LC1	23	20
LC2	21	18
LC3	19	16
PC1	16	16
PC2	17	15
LB1	22	18
LB2	20	17
LB3	22	16



**Fig. 6: Antibacterial activity of CFE of LAB against *L.monocytogenes* by agar diffusion method LB (*Lactobacills*); LC(*Lactococcus*); PC(*Pediococcus*)**

**Table 4: Minimum inhibitory concentration (MIC) of cell free extract of LAB against *L. monocytogenes***

LAB strains	Inhibition percentage (%)		CFE addition volume (ml) to 10 ml of broth
	<i>L.monocytogenes</i> 1	<i>L.monocytogenes</i> 2	
LC 1	43.20	40.13	0.5
LC2	33.85	35.39	
LC3	42.47	40.22	
PC1	28.75	31.53	
PC2	30.10	28.95	
LC1	62.38	67.10	1
LC2	54.47	88.42	
LC3	61.63	95.55	
PC1	55.85	62.24	
PC2	53.42	52.13	
LC1	87.10	98.15	1.5
LC2	92.17	90.78	
LC3	94.24	96.13	
PC1	83.45	89.34	
PC2	89.12	95.92	
LB1	48.97	46.67	0.5
LB2	45.47	42.22	
LB3	46.53	44.70	
LB1	75.69	70.16	1
LB2	71.72	65.56	
LB3	68.48	64.83	
LB1	92.75	82.93	1.5
LB2	94.25	93.69	
LB3	86.94	90.06	

LC(*Lactococcus lactis*), PC(*Pediococcus pentosaceus*), LA(*Lactobacillus plantarum*).

## REFERENCES

- Abd El-Malek, A.M.; Ali1, S.F.H.; Hassanein, R.; Mohamed, M.A.; Elsayh, K.I. (2010). Occurrence of *Listeria species* in meat, chicken products and human stools in Assiut city, Egypt with PCR use for rapid identification of *Listeria monocytogenes*. *J. Vet. World*, **3**(8), 353-359.
- Alsheekh, D.A.A. (1999). Isolation and identification of *lactobacillus acidophilus* and studying of some its properties and using it in production of therapeutically dairy products. Ph.D. Thesis. University of Baghdad.
- Aygun, O.; Pehlivanlar, S. (2006). *Listeria* sp. In raw milk and dairy products in Antakya, Turkey. *J. Food Control*, **17**, 676-679.
- Bille, J.; Rocourt, J.; Swaminathan, B. (1999). *Listeria*, *Erysipelothrix* and *Kurthia*. In Murray, Baron, Pfaller, Tenover and Tenover (ed.). "Manual of Clinical Microbiology". 7th ed. *American Society for Microbiology, Washington, D.C.*
- Brugere-Picoux, O. (2008). Ovine listeriosis. *Small Ruminant Research*, **76**, 12- 20.
- Chukwu, O.O. (2007). Studies of *Listeria monocytogenes* in some selected local government areas of Plateau state, Nigeria. Ph.D. Thesis, University of Jos. Jos.

- Colak, H.; Hampikyan, H.; Bingol, E.B.; Ulusoy, L. (2007). Prevalence of *L. monocytogenes* and *Salmonella* spp. in Tulum cheese. *Food Control, London*, **18**,576-57.
- Dimic, G.R.; Tanackov, S.D.; Jovanov, O.O.; Cvetkovic D.D.; Velicanski, S.L. (2010). Presence of *Listeria* species in meats from rerail markets in Serbia. *Slov. Vet. Research DOI*. **41**, 1-6.
- Ennaji, H.T.; Ennaji, M.; Hassar, M.M.; Cohen, M. (2008). Characterization and antibiotic susceptibility of *Listeria monocytogenes* isolated from poultry and red meat in Morocco. *J. Infection and Drug Resistance*, **1**, 45–50.
- Francoise, L. (2010). Occurrence and role of lactic acid bacteria in seafood products. *J. Food Microbiol.* **27**(6), 698-709
- Fraser, J.A.; Sperber, W.H. (1988). Rapid detection of *Listeria* spp. in food and environmental samples by asculin hydrolysis. *J. Food Prot.*, **38**,1655-1657.
- Glass, K.; Doyle, M.E. (2005). Safety of Processed Cheese: A Review of the Scientific Literature. *Food Research Institute, Madyson*, 1-11.
- Gupta, B.P.; Flores, G.V.; Banerjee, U.; Rodrigues, V. (1998). Patterning an epidermal field: Drosophila lozenge, a member of the AML-1/Runt family of transcription factors, specifies olfactory sense organ type in a dose-dependent manner. *Dev. Biol.*, **203**(2), 400-411.
- Hemeed, A.H. (2004). Using of metabolic products of lactic acid bacteria in preservation of cheese and cream. Ph.D. Thesis. University of Baghdad.
- Jakobsen, R.A.; Heggebo, R.; Sunde E.B.; Skjervheim, M. (2011). *Staphylococcus aureus* and *Listeria monocytogenes* in Norwegian raw milk cheese production. *Food Microbio.*, **28**(3), 492-6.
- Jami, S.; Jamshidi, A.; Khanzadi, S. (2010). The presence of *Listeria monocytogenes* in raw milk samples in Mashhad, Iran. *Iranian J. Vet. Research, Shiraz University*, **11**(4), 363-367.
- Kumar, R. (2011). Modern Trends to Investigate Food Listeriosis. *J. Food Tech.*, **9**(1), 9-17.
- Mansilla, R.T. (2008). Lactic acid bacteria as bioprotective agent against foodborne pathogens and spoilage microorganisms in fresh fruit and vegetable. Ph. D. thesis. Institute of Food and Agricultural Technology, University of Girona ,Spain.
- Marinsek, J.; Grebenc, S. (2002). *Listeria monocytogenes* in minced meat and thermally untreated meat products in Slovenia. *Slovenian Veterinary Research*, **39**(2),131-136.
- Martinez, R.C.; Martinis, E.C. (2006). Effect of *Leuconostoc mesenteroides* 11 Bacteriocin in the multiplication control of *Listeria monocytogenes* 4b. *Cienc. Tecnol. Aliment., Campinas*, **26**(1), 52-55.
- Mead, P.S.; Slutsker, L.; Dietz, V.; McCaig, L.F.; Bresee, J.S.; Shapiro, C.; Griffin, P.M. and Tauxe, R.V. (1999). Food-related illness and death in the United States. *Emerging Infectious Diseases*, **5**, 607-623.
- Nayak, J.B.; Brahmabhatt, M.N.; Savalia, C.V.; Bhong, C.D.; Roy, A.; Kalyani, I.H.; Parmar, B.C. (2010). Detectioin and characterization of *Listeria* species from buffalo meat. *Indian J. Vet. Med.*, **29**(2), 83-94.
- Nespolo, C.R.; Brandelli, A. (2010). Production of bacteriocin –like substance by lactic acid bacteria isolated from regional cheese. *Brazilian J. Microbio.*, **41**, 1009-1018.

- Pattnaik, P.; Sunita, G.; Virender, K.B. (2005). Effect of environmental factors on production of lichenin, a chromosomally encoded bacteriocin-like compound produced by *Bacillus licheniformis* 26L-10/3RA. *Microbiological Research*. **106** (2), 213-218.
- Pereira, C.I.; Gomes, E.O.; Gomes, A.M.P.; Malcata, F.X. (2008). Proteolysis in model Portuguese cheeses: effects of rennet and starter culture. *Food Chem.*, **108**, 862-868.
- Pintado, C.M.B.S.; Olivera, A.; Pampulha, M.E.; Ferreira, M.A.S.S. (2005). Prevalence and characterization of *Listeria monocytogenes* isolated from soft cheese. *Food Microbiol. London*, **22**, 79-85.
- Robert, D.; Greenwood, M. (2003). "Practical in Food Microbiology". 3th ed. Blankwell Publishing Ltd .selar.
- Ross, T.; Dalgaard, P.; Tienungoon, S. (2000). Predictive modelling of the growth and survival of *Listeria* in fishery products. *Int. J. Food Microbiol.*, **62**, 231-245.
- Sammaraco, M.L.; Ripabelli, G.; Fanelli, I.; Grasso, G.M. (2004). Prevalence of *Listeria* spp. in dairy farm and evaluation of antibiotic resistance of isolates. *J. Ann. Ig.*, **17**,175-183.
- Seeliger, H.P.R.; Jones, D. (1987). *Listeria*. In: Holt, J.G. (Ed.), "Bergey's Manual of Systematic Bacteriology". 9th ed. Williams and Wilkins, Baltimore MD.
- Schnurer, J.; Magnusson, J. (2005). Antifungal lactic acid bacteria as biopreservatives. *Trends Food Sci. Technol.*, **16**, 70-78.
- Suskovic, J.; Brkic, B.; Matosic, S.; Maric, V. (1997). *lactobacillus acidophilus* M92 as potential probiotic strain. *Milchwissenschaft*, **52**(8), 430-435.
- Svirakov, E.; Slozilozilova, I.; Tichovsky, P.; Plockova, M. (2009). Effect of *Lactococcus* spp. on the Growth of *Listeria* sp. in the Model UHT Milk System. *Czech J. Food Sci.*, **2**, 8–11.
- Swamina, B.; Gerner-Smidt, P. (2007). The epidemiology of human listeriosis. *Microbes and Infection*, **9**, 1236-1243.
- Swamina, B. (2006). Nationwide Outbreak of Listeriosis due to Contaminated Meat. *Epidemiol. Infect.*, **134**, 744-751.
- Tolga, K.; Gursel, O.; Basak, O.; Ergun, O.G. (2010). Prevalence of *Salmonella* spp. and *Listeria monocytogenes* in different cheese types produced in Turkey. *British Food J.*, **112** (11),1230 – 1236.
- Tompkin, R.B. (2002). Control of *Listeria monocytogenes* in the food-processing. Vazquez-Boland, J.A.; Kocks, C.; Dramsi, S.( 1992). Nucleotide sequence of the lecithinase operon of *Listeria monocytogenes* and possible role of lecithinase in cell-to-cell spread. *Inf. and Immun.*, **60**, 219-230.
- Walter, F. (2000). Food borne Listeriosis. *Clinic. Infect. Disease*. **31**, 770-775.
- Zdolec, N.; Hadziosmanovic, M.; Kozacinski, L.; Cvrtila, Z.; Filipovic, I.; Marcincak, S.; Zeljka, K.; Khalled, H. (2007). Protective effect of *Lactobacillus sakei* in fermented sausages . *Archiv for Lebensmittel hygiene*, **58**, 155–158.