Acta Veterinaria (Beograd), Vol. 49. No. 1, 49–56, 1999.

UDK 619:637.5.06

VARIATIONS IN THE SENSITIVITY OF LISTERIA MONOCYTOGENES TYPES TO LACTIC ACID BACTERIA BACTERIOCINS

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(Received, 5. October 1998.)

Listeria monocytogenes is a cause of human and animal listeriosis, a disease which is often lethal. Food of animal origin is a very important carrier in human infection. This microorganism was found in many foodstuffs during the last decode in numerous studies. As a basic precaution for providing consumer food safety, investigations are directed toward the greatest reduction of the number of undesirable and pathogenic microorganisms in food. Thus, the sensitivity of Listeria monocytogenes, both clinical/human strains (50) and those found in food of animal origin (48), to lactic acid bacteria bacteriocins was examined at two incubation temperatures (37^{0}C/24h and 4^{0}C/12 days). Namely, it is well known that hot only closely velated bacterial species, but also less closely velated Gram positive bacteria, may have bactericidal ov bacteriostatics proteins ov protein molecules to L. monocy togeues. Then they can be eventually added to food, with the aim of decreasing the risk of listeriosis to the minimum. It was found that bacteriocins, originating from Lactococcus UW and Lactobacillus sake 148 did not expressed inhibitory effects on any of the Listeria strains. However, bacteriocins originating from Lactobacillus sake 265, Pediococcus 347 and Lactobacillus sake 706 had listericidal effects on almost every analyzed type. The highest bactericidal effect was expressed by bacteriocins from Lactobacillus sake 265. There was statistically a very significant difference ($p<0.01$) and a significant difference ($p<0.05$) between the arithmetic mean values for inhibition zones with clinical/human strains and those originating from foodstuffs. It was found that the incubation time and temperature influenced the inhibitory effects of bacteriocins, too.

Key words: Listeria monocytogenes, bacteriocins, lactic acid bacteria, temperature treatment, sensitivity.

INTRODUCTION

Direct or indirect contamination of food with many microorganisms, leads to serious disease in humans, which may be lethal (Dimitrijević and Teodorović, 1998). One species which is of current interest, is Listeria monocytogenes, the cause of listeriosis, for which food of animal origin is often a carrier of infection (Farber and Peterkin, 1991; Bunčić, 1991; Bunčić et al., 1995). Great epidemics of listeriosis in humans have occurred after consumption of food of plant origin, milk and dairy products, as well as meat and meat products (Bader, 1993). The ubiquitous nature of Listeria monocytogenes makes very unreal the total exclusion of initial contamination of food (Gahan and Collins, 1991). This indicates the extraordinary importance of knowing well the factors which may reduce its numbers in foodstuffs (Crawford, 1989). Lactic acid bacteria, assigned as health safe microorganisms, are used in food, not only for achieving useful metabolic and sensory changes, but also for their invaluable important protective role, because they have a negative influence on numerous undesirable microorganisms. They achieve their protective role by competition for food and/or by producting bacteriocins and other antimicrobial substances (Klaenhammer, 1993). Bacteriocins are extracellularly released peptides or protein molecules, produced by lactic acid bacteria, which have bactericidal and bacteriostatic effects, usually towards closely related bacterial species (Jack et al., 1995). However, it was found that they may have destructive effects on some unrelated, Gram positive bacterial species among which is L. monocytogenes (Muriana, 1996). Taking in to account that L. monocytogenes is a cause of serious human disease, for which the minimal infective dose is not yet known, and its ubiquitous nature and widespread distribution in foodstuffs it was considered interesting to assess its sensitivity to lactic acid bacteria bacteriocins, which can be eventually added to the food (Eckner, 1992; Teodorović, 1996).

MATERIALS AND METHODS

A collection of 98 strains of L. monocytogenes consisting of 50 clinical/human strains and 48 strains from foodstuffs of animal origin was made. The clinical/human types originated from blood, liquor, infected human feces (carriers or ill persons). Other strains were isolated and identified from foodstuffs of animal origin by the method of McLain and Lee (1989). Bacteriocins from the following lactic acid bacteria were examined: Lactococcus UW, Lactobacillus sake 148, Lactobacillus sake 265, Pediococcus 347 and Lactobacillus sake 706. Assessment of growth inhibition of various types of L. monocytogenes by the bacteriocins was performed by the diffusion method from wells into. Every type was assessed in triplicate in order to provide adequate accuracy and precision of the results. Two incubation procedures were used: 37^0C for 24 hours and 4^0C for 12 days. The results were expressed as the mean values for inhibition zone diameter (mm) for eachstrain The statistical significance of differences between procedures and strain origin calculated by the random plan method using statistical software (Statgraphics 5:0; Statistical Graphic Corporation USA).

Table 1. Sensitivity of L. monocytogenes typs, clinical/human types, related to the bacteriocins of lactic acid bacteria

Sign	Lactobacillus sake 265		Pediococcus 347		Lactobacillus sake 706	
	37°C/24h zone ± SD	4°C/12h zone ± SD	37°C/24h zone ± SD	4°C/12h zone ± SD	37°C/24h zone ± SD	4°C/12h zone ± SD
8	2.00 ± 0.00	10.33 ± 0.57	5.00 ± 0.00	4.00 ± 0.00	1.00 ± 0.00	3.00 ± 0.00
94	2.00 ± 0.00	9.33 ± 0.57	3.00 ± 0.00	3.66 ± 057	1.00 ± 0.00	3.00 ± 0.00
95	4.33 ± 0.28	10.66 ± 0.57	3.66 ± 0.28	4.83 ± 0.76	3.33 ± 0.28	3.83 ± 0.28
96	1.00 ± 0.00	2.00 ± 0.00	2.00 ± 0.00	1.33 ± 0.28	0.50 ± 0.00	1.00 ± 0.00
97	2.66 ± 0.28	5.33 ± 1.15	2.00 ± 0.00	2.33 ± 0.57	0.50 ± 0.00	3.00 ± 0.00
98	1.00 ± 0.00	3.16 ± 0.28	2.66 ± 0.57	2.00 ± 0.00	0.50 ± 0.00	1.33 ± 0.28
99	2.66 ± 0.57	9.00 ± 0.00	4.66 ± 0.57	4.00 ± 1.00	1.50 ± 0.00	2.66 ± 0.57
100	1.00 ± 0.00	9.66 ± 0.57	3.00 ± 0.00	3.00 ± 0.00	1.00 ± 0.00	3.00 ± 0.00
101	1.00 ± 0.00	1.33 ± 0.28	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
102	2.00 ± 0.00	12.33 ± 0.57	4.66 ± 0.57	0	1.00 ± 0.00	3.33 ± 0.57
103	3.33 ± 0.57	9.33 ± 0.57	2.33 ± 0.57	3.33 ± 0.57	1.00 ± 0.00	1.50 ± 0.00
104	1.66 ± 0.28	2.00 ± 0.00	1.16 ± 0.28	2.66 ± 0.28	1.00 ± 0.00	2.00 ± 0.00
105	1.00 ± 0.00	12.66 ± 0.57	4.00 ± 0.00	5.00 ± 0.00	1.00 ± 0.00	4.00 ± 0.00
126	0	3.00 ± 0.00	0	1.00 ± 0.00	0	1.33 ± 0.57
128	1.00 ± 0.00	8.00 ± 1.00	4.00 ± 0.00	3.83 ± 0.28	1.00 ± 0.00	3.16 ± 0.28
132	0	0	0	3.66 ± 0.57	0	3.33 ± 0.28
166	0	3.00 ± 0.00	0	0	0	0
61632	3.00 ± 0.00	9.00 ± 0.00	3.66 ± 0.28	3.00 ± 1.00	1.00 ± 0.00	1.33 ± 0.57
61763	0	0	0	0	0	0
62124	0	6.00 ± 0.00	0	2.83 ± 0.28	0	1.16 ± 0.28
62433	1.66 ± 1.52	5.66 ± 0.57	3.00 ± 0.00	2.33 ± 0.28	1.00 ± 0.00	1.00 ± 0.00
62728	2.00 ± 0.00	9.33 ± 0.57	4.00 ± 0.00	3.83 ± 0.57	1.00 ± 0.00	3.66 ± 0.28
62846	1.00 ± 0.00	9.66 ± 0.57	4.00 ± 0.00	4.00 ± 0.00	1.00 ± 0.00	1.66 ± 0.28
63158	0	1.33 ± 0.28	0	1.33 ± 0.00	0	1.00 ± 0.00
63588	0	2.00 ± 0.00	0.66 ± 1.15	1.50 ± 0.00	1.00 ± 0.00	1.66 ± 0.57
63711	2.00 ± 0.00	6.66 ± 0.57	2.83 ± 0.28	3.00 ± 0.00	1.00 ± 0.00	1.66 ± 0.57
63869	2.66 ± 0.57	4.66 ± 0.57	2.66 ± 1.73	4.66 ± 0.00	1.00 ± 0.00	1.66 ± 0.28
64084	3.00 ± 0.00	7.66 ± 0.57	3.33 ± 0.28	2.16 ± 0.76	1.00 ± 0.00	2.00 ± 0.00
64269	0	1.00 ± 0.00	0	1.00 ± 0.00	0	1.00 ± 0.00
64416	2.66 ± 0.57	8.66 ± 0.57	3.33 ± 0.00	3.00 ± 0.28	1.00 ± 0.00	2.00 ± 0.00
64563	2.00 ± 0.00	2.66 ± 0.28	3.00 ± 0.00	3.00 ± 0.00	2.50 ± 0.00	1.00 ± 0.00
64716	0	7.66 ± 0.57	3.83 ± 0.28	2.50 ± 0.86	0.50 ± 0.00	1.00 ± 0.00
65017	0	10.66 ± 0.57	0	4.00 ± 0.00	0	2.66 ± 0.57
65631	2.66 ± 2.66	6.66 ± 0.57	3.83 ± 0.28	2.00 ± 0.00	0	0.66 ± 0.28
65980	0	1.50 ± 0.50	0	2.33 ± 0.57	0	1.00 ± 0.00
66091	1.00 ± 0.00	2.33 ± 0.57	1.00 ± 0.00	0	0.66 ± 0.28	2.16 ± 0.76
66235	2.83 ± 0.28	7.00 ± 1.00	4.50 ± 0.50	3.83 ± 0.57	1.00 ± 0.00	1.83 ± 0.28
66339	0	7.33 ± 0.57	0	3.00 ± 0.00	0	2.50 ± 0.00
66575	0	3.00 ± 0.00	0	1.66 ± 0.57	0	1.66 ± 0.28
66824	3.00 ± 0.00	6.00 ± 0.00	3.83 ± 0.28	3.83 ± 0.28	1.83 ± 0.28	2.83 ± 0.28
67143	3.00 ± 0.00	2.33 ± 0.57	1.83 ± 0.28	0	1.83 ± 0.28	0.50 ± 0.00
67324	2.66 ± 0.28	9.00 ± 0.00	4.00 ± 0.00	3.00 ± 0.00	1.00 ± 0.00	2.66 ± 0.28
76479	2.66 ± 0.28	10.66 ± 0.57	4.33 ± 0.57	3.66 ± 0.57	1.66 ± 0.28	3.83 ± 0.28
67675	2.00 ± 0.00	7.33 ± 0.57	4.00 ± 0.00	2.00 ± 0.00	1.00 ± 0.00	1.50 ± 0.00
67977	0	1.00 ± 0.00	0	1.16 ± 0.28	0	1.00 ± 0.00
67992	0	3.00 ± 0.00	0	1.66 ± 0.57	0	1.00 ± 0.00
68602	2.00 ± 0.00	7.00 ± 0.57	3.00 ± 0.00	2.16 ± 0.28	0	1.50 ± 0.00
68614	0	4.00 ± 0.00	0	2.00 ± 0.00	0	1.00 ± 0.00
69300	3.00 ± 0.00	0	3.33 ± 0.57	4.00 ± 0.00	1.00 ± 0.00	2.33 ± 0.57
69873	0	0	0	0	0.50 ± 0.00	1.00 ± 0.00
Br. + s. (%)	33 (66)	46 (92)	35 (70)	44 (88)	34 (68)	48 (96)
Xsr. ± SD	2.16 ± 0.90	6.14 ± 3.43	3.14 ± 1.18	2.77 ± 1.11	1.11 ± 0.57	1.95 ± 0.99
BR. - s. (%)	17 (34)	4 (8)	15 (30)	6 (12)	16 (32)	2 (4)

Br. + s. (%)... number of positive types L. monocytogenes (percent)
Xsr. mean value of inhibition zone
Br. - s. (%) ... numbres of negative types L. Monocytogenes (percent)
SD ... standard deviation

Table 2. Sensitivity of L.monocytogenes types, types from foodstuff, related to the bacteriocins of lactic acid bacteria

Sign	Lactobacillus sake 265		Pediococcus 347		Lactobacillus sake 706	
	37°C/24h zone ± SD	4°C/12h zone ± SD	37°C/24h zone ± SD	4°C/12h zone ± SD	37°C/24h zone ± SD	4°C/12h zone ± SD
48	2.16 ± 0.28	633. ± 0.57	3.83 ± 0.28	2.66 ± 0.28	1.00 ± 0.00	2.00 ± 0.00
53	2.00 ± 0.00	1.66 ± 0.28	1.66 ± 0.28	2.66 ± 0.28	1.00 ± 0.00	1.00 ± 0.00
54	1.83 ± 0.28	3.00 ± 0.00	1.66 ± 0.28	2.33 ± 0.28	1.00 ± 0.00	1.33 ± 0.28
55	2.00 ± 0.00	5.33 ± 0.57	3.66 ± 0.28	2.00 ± 0.00	1.00 ± 0.00	1.66 ± 0.28
56	0.50 ± 0.86	3.00 ± 0.00	0	1.66 ± 0.57	1.00 ± 0.00	1.16 ± 0.28
57	2.00 ± 0.00	8.00 ± 0.00	3.00 ± 0.00	4.66 ± 0.57	1.00 ± 0.00	4.33 ± 0.57
58	8.33 ± 0.57	0	0.66 ± 1.15	0	1.83 ± 0.28	0
59	3.00 ± 0.00	3.00 ± 0.00	1.16 ± 0.28	2.00 ± 0.00	1.16 ± 0.28	1.16 ± 0.28
60	2.00 ± 0.00	3.50 ± 0.50	3.00 ± 0.50	2.33 ± 0.28	2.00 ± 0.00	1.50 ± 0.00
61	2.66 ± 0.57	0	1.50 ± 0.50	0	1.00 ± 0.00	0.33 ± 0.28
62	2.00 ± 0.00	2.00 ± 0.00	1.00 ± 0.00	2.00 ± 0.00	1.16 ± 0.28	1.00 ± 0.00
63	2.83 ± 0.28	7.66 ± 0.57	4.00 ± 0.00	4.33 ± 0.57	2.00 ± 0.00	2.50 ± 0.00
66	0	4.33 ± 0.57	0	2.16 ± 0.28	0	1.66 ± 0.28
67	1.33 ± 0.28	3.66 ± 0.57	3.33 ± 0.57	2.33 ± 0.57	1.00 ± 0.00	2.00 ± 0.00
68	4.00 ± 0.00	4.83 ± 0.28	3.00 ± 0.00	2.00 ± 0.00	1.83 ± 0.28	1.00 ± 0.00
69	0	9.66 ± 0.57	5.00 ± 0.00	5.33 ± 0.28	3.33 ± 0.57	4.00 ± 0.00
70	0	5.00 ± 0.00	0	1.66 ± 0.28	0	1.50 ± 0.00
71	2.50 ± 0.50	4.66 ± 0.57	3.33 ± 0.57	2.00 ± 0.00	1.00 ± 0.00	2.00 ± 0.00
72	1.50 ± 0.00	5.33 ± 0.57	3.66 ± 0.57	2.66 ± 0.28	0.50 ± 0.00	1.66 ± 0.28
74	1.16 ± 0.28	5.00 ± 0.00	2.33 ± 0.28	3.83 ± 0.28	1.00 ± 0.00	2.66 ± 0.57
77	1.16 ± 0.28	3.83 ± 0.28	2.66 ± 0.57	2.00 ± 0.00	1.00 ± 0.00	2.33 ± 0.57
78	0	0	0	0	0	0
79	0	0	0	0	1.00 ± 0.00	1.50 ± 0.00
82	0	4.00 ± 0.00	0	2.50 ± 0.00	0	2.16 ± 0.28
83	1.50 ± 0.00	5.00 ± 0.00	3.00 ± 0.00	2.50 ± 0.00	4.00 ± 0.00	2.50 ± 0.00
84	2.00 ± 0.00	2.00 ± 0.00	2.66 ± 0.28	1.66 ± 0.28	1.00 ± 0.00	1.66 ± 0.28
90	1.66 ± 0.28	8.33 ± 0.57	3.33 ± 0.57	3.00 ± 0.00	0	2.00 ± 0.00
91	1.50 ± 0.00	3.66 ± 0.57	2.66 ± 0.28	2.00 ± 0.00	1.00 ± 0.00	2.00 ± 0.00
92	1.66 ± 0.28	2.66 ± 0.57	2.00 ± 0.00	1.83 ± 0.28	1.33 ± 0.28	2.00 ± 0.00
125	1.50 ± 0.00	3.00 ± 0.00	2.33 ± 0.28	2.00 ± 0.00	1.00 ± 0.00	2.16 ± 0.28
127	2.66 ± 0.57	8.66 ± 0.57	4.83 ± 0.28	1.00 ± 0.00	1.33 ± 0.28	2.00 ± 0.00
130	0	0	0	0	0	0
134	1.66 ± 0.28	4.00 ± 0.00	2.83 ± 0.28	1.83 ± 0.28	1.00 ± 0.00	1.00 ± 0.00
157	1.00 ± 0.00	5.00 ± 0.00	3.83 ± 0.28	1.66 ± 0.28	0	1.66 ± 0.28
168	0	0	0	0	0	0
169	0.50 ± 0.00	0	1.00 ± 0.00	1.00 ± 0.00	0	1.00 ± 0.00
170	0	0	0	0	0	0
171	1.00 ± 0.00	4.66 ± 0.28	3.66 ± 0.57	1.00 ± 0.00	0	0
70172	1.50 ± 0.50	4.66 ± 0.57	3.00 ± 0.00	1.83 ± 0.28	0.83 ± 0.28	1.16 ± 0.28
70313	3.00 ± 0.00	0	1.00 ± 0.00	0	1.16 ± 0.28	0
70550	2.33 ± 0.57	9.00 ± 0.00	1.66 ± 0.28	3.33 ± 0.57	1.66 ± 0.28	2.33 ± 0.57
70592	2.00 ± 0.00	2.66 ± 0.28	1.50 ± 1.32	1.50 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
70620	0	3.83 ± 0.28	0	1.33 ± 0.57	0	1.00 ± 0.00
70708	2.00 ± 0.00	3.66 ± 0.57	3.33 ± 0.57	1.00 ± 0.00	1.00 ± 0.00	1.00 ± 0.00
70722	1.00 ± 0.00	7.33 ± 0.57	2.00 ± 0.00	2.00 ± 0.00	0.50 ± 0.00	2.33 ± 0.28
70841	0	7.66 ± 0.57	0	2.33 ± 0.57	0	2.00 ± 0.00
70916	0	5.00 ± 0.00	0	2.50 ± 0.00	0	2.50 ± 0.28
71062	2.00 ± 0.00	2.00 ± 0.00	1.00 ± 0.00	1.66 ± 0.28	1.00 ± 0.00	1.00 ± 0.00
Br. + s. (%)	36 (75)	39 (81.25)	36 (75)	40 (83.33)	34 (70.83)	41 (85.41)
Xsr. ± SD	2.04 ± 1.31	4.66 ± 2.22	2.61 ± 1.16	2.25 ± 0.97	1.29 ± 0.72	1.77 ± 0.79
Br. – s. (%)	12 (25)	9 (18.75)	12 (25)	8 (16.66)	14 (29.16)	7 (14.58)

RESULTS AND DISCUSSION

Undeniably some bacteriocins originating from lactic acid bacteria may have bactericidal or bacteriostatic effects, towards both closely realted bacterial species, and also towards less closelly related Gram positive species such as L. Monocytogenes (Dimitrijević, 1998). In our study it was found that those bacteriocins originating from the chosen lactic acid bacteria did not express an inhibitory effect on the tested types of L. monocytogenes in all cases (Table 1 and 2).

Table 3. Statistical significance of differences beetween mean inhibition zones for clinical/human types of L.monocytogenes and types from foodstuff after 24 hours at 38°C in the presence of three bacteriocins

	X	Lb. 706 clin	Lb. 706 food	Lb. 265 clin	Lb. 265 food	Ped. 347 food	Ped. 347 clin
Ped.347 clin	2.20	1.44**	1.29**	0.77**	0.66**	0.23	–
Ped.347 food	1.96	1.20**	1.05**	0.53**	0.43**	–	–
Lb.265 food	1.53	0.77**	0.62**	0.10	–	–	–
Lb.265 clin	1.43	0.67**	0.52**	–	–	–	–
Lb..706 food	0.90	0.15	–	–	–	–	–
Lb.706 clin	0.75	–	–	–	–	–	–

** $p<0.01$ (significance at 99%)

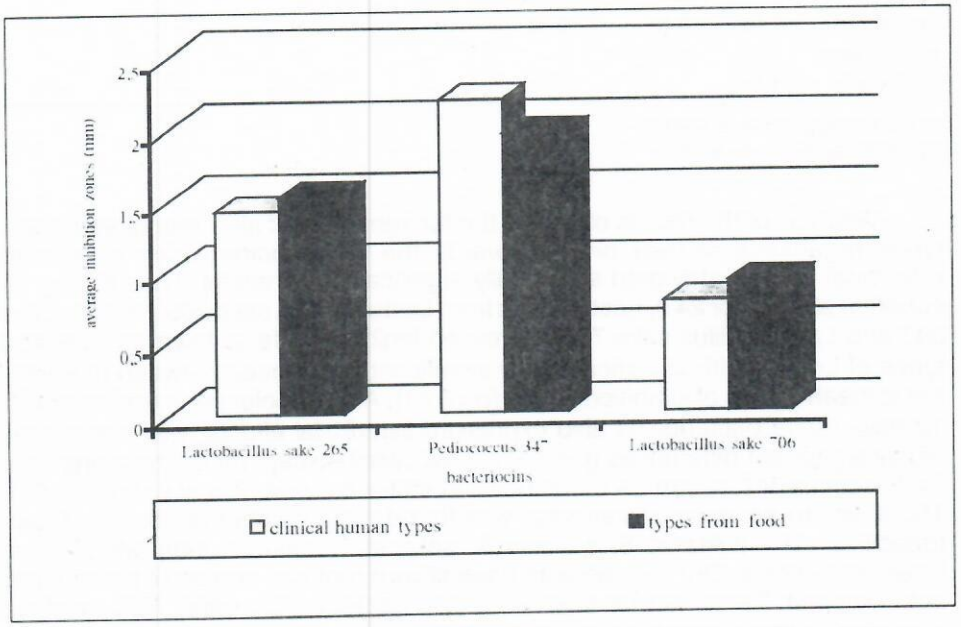

Figure 1. Sensitivity variations of L. monocytogenes types to lactic acid bacteria bacteriocins at 37°C/24h

Bacteriocins from Lactococcus UW and Lactobacillus sake 148 did not express bactericidal activity in any case. Bacteriocins from Lactobacillus sake 265,Pediococcus 347 and Lactobacillus sake 706 expressed bactericidal effects on all except a small number of tested strains of L. monocytogenes. It was found that there was no statistically significant difference between the arithmetic mean values for inhibition zones, with clinical/human types and types from foodstuffs after 24 hours at 37^{0}C, (Table 3, Figure 1.) in relation to the assessed bacteriocins ($p<0.05$). However, at a cooler temperature (4^{0}C) for 12 days, it was found that there were statistically significant, differences in the inhibitory effects of bacteriocins originating from Lactobacillus sake 265, and Pediococcus 347, in favor of clinical/human types (Table 4, Figure 2). On this basis it can be claimed, with 99% ($p<0.01$), and 95% ($p<0.05$) probability that,bacteriocins originating from lactobacillus sake 265, and Pediococcus 347, respectively will express a greater inhibitory effect towards clinical/human types than towards types originating from foodstuffs of animal origin.

Table 4. Statistied significance of differences between mean inhibition zones for clinical/human types of L.monocytogenes and types from foodstuff after 12 days at 4^{0}C in the presence of three bacteriocins

	$\bar{X}$	Lb. 706 clin	Lb. 706 food	Lb. 265 clin	Lb. 265 food	Ped. 347 food	Ped. 347 clin
Ped.265 clin	5.65	4.13**	3.77**	3.77**	3.21**	1.76**	–
Ped.265 food	3.88	2.37**	2.01**	2.00**	1.44**	–	–
Lb.347 clin	2.44	0.92**	0.56*	0.56*	–	–	–
Lb.706 clin	1.88	0.36	0.00	–	–	–	–
Lb..347 food	1.87	0.36	–	–	–	–	–
Lb.706 clin	1.51	–	–	–	–	–	–

** $p<0.01$ (significance at 99%)
* $p<0.05$ (significance at 95%)

Analysis of the results obtained the for sensitivity of all L. monocytogenes, types regardless of their origin, towards the bacteriocins which expressed listericidal effects, indicated statistically significant differences. Thus during incubation at 37^{0}C for 24 h, bacteriocins from Lactobacillus sake 265, Pediococcus 347 and Lactobacillus sake 706 expressed inhibitory effects towards all tested types of Listeria with statistically very significant differences between the arithmetic mean values of inhibition zones ($p<0.01$). At the cooler temperature of 4^{0}C 12 days these bacteriocins also expressed listericidal effects, with statistically highly significant differences ($p<0.01$), in all cases, except for bacteriocins from Pediococcus 347, where the difference was statistically significant only ($p<0.05$). The ability to produce bacteriocins was found for a number of strains of Lactobacillus sake (laktacin S, sakacin A, sakacin P, sakacin 148) which have listericidal effects. Our experiments have shown that two strains of these lactic acid bacteria (Lactobacillus sake 265, and Lactobacillus sake 706) produce bacteriocins that have inhibitory effects on the assessed Listeria. The effects were greater at a cool temperature over a long period. Bacteriocins originating from

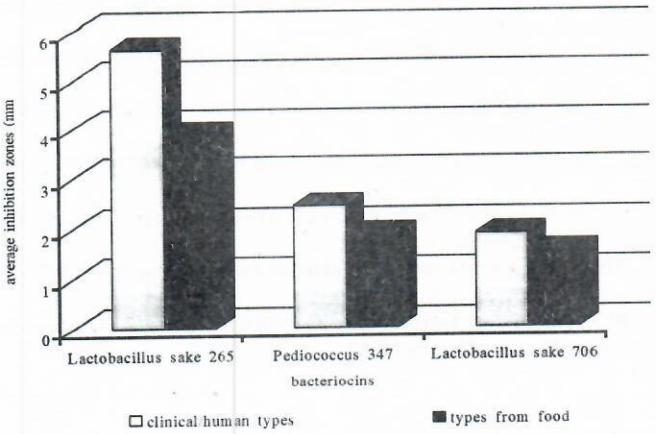

Figure 2. Sensitivity variations of L. monocytogenes types to lactic acid bacteria bacteriocins at 4^0C/12 days

Lactobacillus sake 265 gave much greater inhibition zones (1.43-5.65 mm) than bacteriocins originating from Lactobacillus sake 706 (0.76-1,88), which had the lowest listericidal effect. Pediococcus 347 expressed a very uniform listericidal effect (1.88-2.44 mm) both at the cool temperature (4^0 C for 12 days) and at 37^0C for 24 h. However, it was suggested that pediococci also produce some acids, as metabolic products, which might inhibit the growth of Listeria (Raccach and Geshel, 1993), but we excluded that possibility by using separated bacteriocin. On the basis of the data presented the addition of lactic acid bacteria bacteriocins to food should be considered in the future, with the aim of reducing the eventual presence of Listeria in foodstuffs of animal origin and therefore lowering the risk of alimentary listeriosis to a minimum.

REFERENCES

1. *Bader J.* 1993. Listeriosis epidemic. *Lancet, 342,* 607.
2. *Bunčić S.* 1991. The incidence of Listeria monocytogenes in slaughtered animals, in meat, and in meat products in Yugoslavia.Int J. Food. Microbiol., 12, 173-180.
3. *Bunčić S., Baltić M., Panin J., Tadić I., Teodorović V.* 1995. Noviji naučni izazovi u oblasti higijenske ispravnosti i kvaliteta mesa i proizvoda od mesa. *Tehnologija mesa, 2-3,* 146-157.
4. *Crawford L. M.* 1989. Revised policy for controlling *Listeria monocytogenes. Fed regist., 54,* 22345-22346.
5. *Dimitrijević M.* 1998. Ispitivanje varijacija između sojeva Listeria monocytogenes u pogledu osetljivosti prema baktericionima mlečnokiselinskih bakterija. Magistarska teza, Beograd.
6. *Dimitrijević M., Teodorović V.* 1998. Značaj namirnica animalnog porekla u nastanku alimentarne listerioze. Vet. Glasnik, vpl. 52, 1-2, str. 1-128, 79-85

7. *Eckner K. F.* 1992. Bacteriocins and food applications. Dairy Food Environ. Sanit., 12, 204-209.
8. *Farber J. M., Peterkin P. I.,* 1991. Listeria monocytogenes, a foodborne pathogen. *Microbiol. Rev., 55,* 476-511.
9. *Gahan C. G. M., Collins J. K.* 1991. Listeriosis: biology and implications for the food industry. *Trends Food Sci. Technol., 2,* 89-93.
10. *Jack R. W., Tagg J. R., Ray B.* 1995. Bacteriocins of gram-positive bacteria. *Microbiol. Rev., 59,* 171-200.
11. *Klaenhammer T. R.,* 1993. Genetics of bacteriocins produced by lactic acid bacteria. FEMS Microbiol. Rev., 12, 39-86.
12. *Muriana P. M.* 1996. Bacteriocins for control of Listeria spp. in food J. Food Protect. Supplement, 54, 54-63.
13. *Raccach M., Geshell D. J.* 1993. The inhibition of Listeria monocytogenes in milk by pediococci. Food *Microbiol., 10,* 181-186.
14. *Teodorović V.,* 1996. Mogućnost korišćenja bakteriocina u industriji mesa. *Tehnologija mesa, 3-4,* 173-176.
15. *McLain D., Le W. H.* (1989). Isolation and identification of Lutera monocytagens from moat. USDA (FSLS) *Microbiol.* Dcu. Lb. Comm. N° 57

VARIJACIJE OSETLJIVOSTI SOJEVA LISTERIA MONOCYTOGENES PREMA BAKTERIOCINIMA MLEČNOKISELINSKIH BAKTERIJA

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SADRŽAJ

Listeria monocytogenes je uzročnik listerioze ljudi i životinja, oboljenja koje se često završava letalno. Namirnice animalnog porekla su veoma važan vektor u nastanku infekcije ljudi. Tokom poslednje dekade mnogobrojnim istraživanjema ustanovljeno je prisustvo ovog, inače ubikvitarnog mikroorganizma, u velikom broju namirnica. Kako je higijenska ispravnost namirnica animalnog porekla osnovni preduslov za obezbeđivanje zdravstvene bezbednosti i sigurnosti potrošača, nastojanja su usmerena na što većoj redukciji prsustva nepoželjnih i patogenih mikroorganizama u hrani. U ovom radu ispitivana je osetljivost sojeva L. monocytogenes, kliničkih/humanih (50) i sojeva poreklom iz nairnica (48) prema baktericinima mlečnokiselinskih bakterija, pri dve temperature inkubacije(37^{0}C/24h) i 4^{0}C/12 dana). Naime, poznato je baktericidno i bakteriostatsko dejstvo ovih proteina ili proteinskih molekula ne samo prema srodnim bakterijskim vrstama, već i prema manje srodnim Gram pozitivnim bakterijama, kao što je L. mnocytogenes, te bi se eventualno mogli dodavati u hranu, u cilju smanjenja rizika od listerioze na minimum. U radu je ustanovljeno da bakteriocini poreklom od Lactococcus UW i Lactobacillus sake 148 nisu ispoljili inhibitorni efekat ni prema jednom soju listerija, dok su bakteriocini poreklom od Lactobacillus sake 265, Pediococcus 347 i Lactobacillus sake 706 imali listericidni efekat skoroprema svim ispitivanim sojevima. Najveće baktericidno dejstvo ispoljio je bakteriocin poreklom od Lactobacillus sake 265. Postajala je statistički vrlo značajna ($p<0.01$), odnosno značajna razlika ($p<0.05$) između aritmetičkih sredina zona inhibicije kliničkih/humanih i sojeva poreklom iz namirnica. Takođe ustanovljeno je da je temperatura inkubacije uticala na inhibitorne efekte bakteriocina.