

SUMMARY

Listeria monocytogenes are Gram-positive, relatively anaerobic, non-spore-forming rods. These bacteria show the ability to adapt to adverse conditions. In the food processing environment, *L. monocytogenes* are exposed to various stress factors. This stress can be lethal, causing irreversible damage to the cell, or sublethal, i.e., allowing survival.

The purpose of this dissertation was to evaluate the effects of stress factors on changes in pheno- and genotypic characteristics of *L. monocytogenes* strains.

Twenty isolates each from clinical material, salmon, cured meats and frozen vegetables, respectively, were studied. It has been distinguished fifty genetically different strains of *L. monocytogenes*. Sixty-two % of the strains represented serogroup 1/2a-3a, and 60.0% had ten virulence genes (*fbpA*, *plcA*, *hlyA*, *plcB*, *inlB*, *actA*, *iap*, *inlA*, *mpl*, *prfA*). The majority (46; 92.0%) of the strains were susceptible to the tested antibiotics.

In the next step, the tested strains were subjected to selected stresses: osmotic, acidic and alkaline, high and low temperature, cyclic freezing and defrosting (4 cycles), high and low nutrient availability. The median growth-inhibiting concentration of NaCl for strains from all groups by origin was 8.0%. The median growth inhibitory value for pH < 7.0 was 5.0, 4.5, 4.0 and 3.5 for strains isolated from cold cuts, clinical material, frozen foods salmon, respectively. The median growth inhibitory value for pH > 7.0 was 11.0 for clinical strains and 9.0 for strains isolated from cold cuts, frozen vegetables and salmon. There were no statistically significant differences in the response to osmotic, acid and alkaline stress depending on the origin of the strains. Five strains survived a 60-minute exposure to 70°C. All strains survived all stages of the cold stress experiment. During the freezing-defrosting cycle the bacteria number increased after the first cycle. There were no significant differences in the bacteria number between the variant of 4 cycles of freezing-defrosting and the variant of defrosting after the fourth cycle. The survival of the tested strains was demonstrated under varying nutrient availability, both excess and

deficiency. The greatest resistance to stress factors was shown by strains isolated from clinical material. The least tolerant were strains isolated from cold cuts and frozen foods.

Further experiments were conducted on two strains and the reference strain *L. monocytogenes* ATCC 19111. The effect of selected stresses: heat, cold, osmotic, acid, alkali, frozen on phenotypic features: MIC (minimum inhibitory concentration) of antibiotics, motility and ability to form a biofilm and expression level of *sigB*, *agrA* genes has been assessed. Variations in the MIC values of the antibiotics included in the study were demonstrated, and these variations were strain and stress factor dependent. Strain 472W formed a strong biofilm after exposure to most stress factors, with the exception of heat and acid stress. Strain 55K, formed a strong biofilm after exposure to low temperature stress. Increased levels of *sigB* gene expression were found in acid stress (strains 55K and 472W) and heat stress (strain 472W). In contrast, increased levels of *agrA* gene expression were recorded under alkali stress (strain 55K). Nonetheless, there was no link between the biofilm formation ability and *sigB* and *agrA* expression after exposure to stress.

L. monocytogenes are exposed to many different stress factors in the food industry and in the environment. It has been shown that the response to both sublethal and lethal conditions varies and depends on strain. Knowledge about the stress response of *L. monocytogenes* will allow the planning of appropriate disinfection strategies to reduce the spread of these pathogens in healthcare buildings, public buildings, and food processing environments.

Keywords: *Listeria monocytogenes*, environmental stress factors, biofilm, antibiotic resistance, gene expression