



Response of *Listeria* to stress

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A total of 173 *Listeria monocytogenes* strains were isolated from humans, animals, the environment and food and were analyzed for the presence of plasmids (Lebrun *et al.*, 1992). Plasmids were found in 28% of the isolates and were more frequently extracted from serogroup 1 strains (35%) than from serogroup 4 strains (15%). Among isolates from food and the environment, 40% and 29%, respectively, harboured plasmids, whereas only 13% of the strains from humans and animals with listeriosis harbored plasmids. Among the 48 plasmid-bearing strains, only 1 harboured 2 plasmids. On the basis of the numbers and sizes of the plasmids, 19 plasmid profiles were defined. The susceptibility of 90 strains to 7 antibiotics and 4 heavy-metal salts was investigated. A total of 95.3% of the plasmid-positive strains and only 12.7% of the plasmid-negative strains were resistant to cadmium. Some 297 *L. monocytogenes* strains isolated from humans and the environment were analyzed for cadmium susceptibility and 44.8% were found to be resistant to cadmium (MICs > 16 µg/ml). Cadmium resistance was found more frequently in serogroup 1 strains (50.9%) than in serogroup 4 (27.8%). The relationships between plasmids or cadmium resistance and phage typing results were investigated (Audurier and Martin, 1989). Plasmids were more common among non-phage-typable strains (38.2%) than among phage-typable strains (22%). Loss or gain of plasmid resulted in phage-type variations. The loss of cadmium resistance in strains harbouring plasmids was obtained for 20 serogroup 1 isolates by high-temperature treatment. Phage typing results of cadmium-susceptible and cadmium-resistant isogenic variants were compared. Of the 88 lytic reactions obtained, 52.3% were unchanged with the loss of plasmid. Seven phages did not

show any lytic reaction. For the further 14 phages, lytic reaction variability with the loss of plasmid fluctuated between 21 and 100%. None of these 14 phages yielded a total reproducibility. The phage-type variations consisted in 27 losses and 15 gains in lytic reactions. In conclusion, (i) plasmid profile analysis might be used in non-phage-typable *L. monocytogenes* strains, and (ii) loss or gain of plasmids in *L. monocytogenes* strains results in phage-type variation.

References

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Response of *Listeria* to stress. T Gormon, L Phan-Thanh (INRA-Tours, Laboratoire de Pathologie Infectieuse et Immunologie, 37380 Nouzilly, France)

Stress provokes changes in the macromolecular composition and structural organization of the cell. The change in individual proteins is of particular importance, since many proteins are involved in numerous enzymatic reactions of the metabolism of the microorganism. Stressing agents are common and diverse in nature. The proteins induced in *Listeria* by low (4°C) and high (49°C) temperatures, extreme pH (pH 4 and pH 9.5), detergents (0.015% SDS, 0.3% deoxycholate) and ethanol (5%) were analyzed using bidimensional gel electrophoresis and a computer-aided 2D analysis system. Stress repressed approximately half the number of proteins synthesized under normal conditions and decreased the level of many others. Conversely, the syn-

thesis of a great number of proteins was enhanced and novel proteins appeared upon stress. Each stress induced a set of specific proteins. However, there were overlaps between these sets of specific proteins. This suggests that those common proteins may be the gene products of common inducible regulating genes in a multigene system of regulation with multiple signals. Furthermore, the same stress induced a certain number of common proteins in 2 different species of *Listeria* (*L. monocytogenes* and *L. innocua*). The most prominent ubiquitous stress protein of *Listeria* had a molecular weight of 17.6 and a pI of 5.1. This protein was found to be induced in every stress investigated and with an induction ratio from 5- to 50-fold depending on the nature of stress. These facts suggest that related or similar bacteria may develop similar (if not the same) strategies (mechanisms) in response to the same stress.

Prediction of *Listeria* development on meat surface. JD Daudin, I Desnier, J Labadie, C Laplace, A Lebert (*INRA-Theix, Station de Recherches sur la Viande, 63122 Saint-Genès-Champanelle, France*)

Chilling and storage conditions influence the development of *Listeria* on meat surfaces. The effects of main abiotic factors were studied *in vitro*. *Listeria* strains isolated from meat (43 *L. monocytogenes* and 8 *L. innocua*) were cultured in phosphate-buffered tryptic meat broth (TMB), whose composition is close to that of meat. NaCl was added to adjust the water activity (A_W). Growth was followed by optical density (OD) measurements with a photometer using microplates (Bioscreen C, Labsystem, Finland). Each factor (temperature, pH, A_W) was tested at 2 levels (low and high) and the number of experiments was reduced by using a Tagushi design. Four conditions were tested: 37°C, A_W 0.96, pH 5.6; 37°C, A_W 1, pH 7.0; 10°C, A_W 0.96, pH 7.0; 10°C

A_W 1, pH 5.6. Growth curves were fitted by a Gompertz function:

$$f(t) = A \cdot \exp \left[- \exp \left[\frac{\mu e}{A} (L - t) + 1 \right] \right]$$

and growth parameters, lag time (L), maximal growth rate (μ) and maximal population level (A), were determined. Optical density is related to bacteria concentration, growth curves were expressed as $\log(\text{OD}/\text{OD}_{\text{at } t=0}) = f(t)$. The Gompertz function fitted the curves well. The generation time at 10°C was 13 ± 3 h, and the lag time varied from 17 ± 7 h at A_W 1 to 32 ± 19 h at A_W 0.96. The conditions at 10°C are the most disadvantageous and larger differences can be observed between strains. At 37°C, the generation time was 0.7 ± 0.1 h and the lag time was 0.9 ± 0.6 h in the best conditions. When both A_W and pH were low, the generation time was 1.7 ± 0.3 h and the lag time was 4.0 ± 2.1 h. No correlation was observed between the growth parameters and the strain serotype, *L. innocua* were characterized by higher lag time and maximum population than *L. monocytogenes*. The differences between strains must be considered in the chilling and storage conditions of meat. This study permitted the beginning of a greater project (Usine Ultra-propre) which concerns the testing of more strains and the selection of some of them to study their growth on meat surfaces.

Factors affecting growth of *Listeria monocytogenes* on fresh, ready-to-use salads. F Carlin, C Nguyen-The (*INRA, Domaine Saint-Paul, 84143 Montfavet Cedex, France*)

Listeria monocytogenes was isolated from fresh, ready-to-use salads with frequencies varying from 0 to 19% (Lainé and Michard, 1988; Velani and Roberts, 1991; Beaufort *et al*, 1992). Concern was expressed that