CELL ADAPTATION OF *Tetragenococcus halophilus* FOR HEAT TOLERANCE

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ABSTRACT

Lactic acid bacteria (LAB) are commonly used as starter cultures in food fermentation. Lactic acid produced by LAB could cause an acidic condition and is lethal to most food spoilage bacteria and food poisoning bacteria. *Tetragenococcus halophilus* (a type of LAB) is frequently found in high-salt fermented food products such as soy sauce. It is a promising LAB to be used as commercial starter culture. A spray-drying method can be employed to produce this culture in powder form, resulting in low cost transportation and prolonged storage. However, the obvious challenge associated with using spray drying is the viable loss of bacteria after heat treatment. Understanding the mechanism of stress response from salt and heat of LAB may lead to the development of cultures with improved capacity to survive and function under industrial production conditions. Therefore, this study determined the effects of salt and heat on cell adaptation of *T. halophilus* which may lead to cell viability after heat exposure in spray drying. When *T. halophilus* were cultured in MRS broth with (3, 5, 10, 15 and 18% NaCl) and without (0% NaCl) the addition of sodium chloride at 37°C for 24 h, the highest growth was found in MRS broth containing 10% NaCl (10-MRS) at 37°C, compared to those in 10-MRS at 30, 42 and 45°C for 48 h. Cell adaptation of *T. halophilus* to salt (10-MRS to 18-MRS) and to heat (37°C to 42°C) for 36 h gave higher survivors and D-values as compared to those from non-adapted cultures, after heat treatment in 0.1M phosphate buffer at 50°C for 3 h. The heat response proteins of *T. halophilus* in salt or heat or in salt and heat combined adaptation for 0, 12, 24 and 36 h were demonstrated by proteomics approaches. Protein identification by LC-MS/MS revealed the differential expression of general stress proteins (DnaK and chaperonin 60), proteins involved in energy metabolism (enolase, acetate kinase and FpF1-ATPase) and proteins related to translation processing (translation elongation factor 1A) after *T. halophilus* adapted to salt, heat and combination of salt and heat. The ATP-dependent Clp protease, ATP-binding subunit ClpE was uniquely expressed after heat adaptation for 12 h. Moreover, proteins involved in cell envelope synthesis, central intermediary metabolism, fatty acid and phospholipids metabolism, purines, pyrimidines, nucleosides and nucleotides biosynthesis and regulatory functions were specifically expressed in cell adapted to combination of salt and heat for 36 h. Finally, the unknown function proteins were also detected, and these proteins might provide the discovery of potential novel biomarker involved in heat tolerance of *T. halophilus* in the future.

KEY WORDS: *Tetragenococcus halophilus* / HEAT SHOCK PROTEINS / SALT AND HEAT STRESSES / LC-MS/MS

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