Journal of Applied Microbiology 2000, **88**, 877–886

Isolation and characterization of a new bacteriocin from *Lactobacillus gasseri* KT7

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7432/10/99: received 15 October 1999, revised 14 January 2000 and accepted 18 January 2000

W.M. ZHU, W. LIU AND D.Q. WU. 2000. A bacteriocin-producing *Lactobacillus gasseri* strain, KT7, was isolated from infant faeces. The supernatant fluid showed inhibitory activity not only against some lactic acid bacteria but also, against some pathogenic and food-spoilage species, including *Clostridium*, *Listeria* and *Enterococcus*. An antimicrobial peptide designated gassericin KT7 was isolated from *Lactobacillus gasseri* KT7. It was purified to homogeneity by a single four-step procedure: a crude supernatant fluid obtained from early stationary-phase culture in MRS medium was subjected to ammonium sulphate fractionation, CM-Sephadex cation-exchange chromatography, Phenyl-Sepharose hydrophobic chromatography and reverse-phase HPLC chromatography. Gassericin KT7 was sensitive to proteolytic enzymes, resistant to heat, active over a wide range of pH, and migrated as a 4.5–5.0 kDa peptide on SDS–PAGE. The bacteriocin was produced constitutively during exponential growth. It was bactericidal to sensitive cells and the bactericidal effect was not produced by cell lysis. The amino acid composition of the bacteriocin was determined and no modified amino acid was found among the residues identified.

INTRODUCTION

Lactic acid bacteria (LAB) are widely used for the fermentation and preservation of a wide range of milk, meat and vegetable foods (Daeschel 1989). The antimicrobial compounds produced by these bacteria include organic acids, hydrogen peroxide, diacetyl and bacteriocins and play an essential role in ensuring the safety and extending the shelf-life of these products. Antimicrobial activities of LAB have been widely investigated in the past 20 years (Jack *et al*. 1995). Increasing consumer demand for 'natural' and 'additive-free' products has led to greater interest in the application of natural inhibitory substances as food preservatives, which could replace or reduce the use of chemical additives. Lactic acid bacteria bacteriocins are biologically-active proteins or protein complexes that act bactericidally against Gram-positive bacteria, usually closely related to the producer strain (Klaenhammer 1988). Bacteriocin production is a common phenomenon among LAB. It has been observed among *Lactobacillus*, *Lactococcus*, *Pediococcus*, *Leuconostoc* and *Carnobacterium* species (Geis *et al*. 1983; Orberg and Sandine 1984; Daeschel and Klaenhammer 1985; Bhunia *et al*. 1988; Spelhaug and Harlander 1989; Ahn and Stiles 1990; Rammelsberg and Radler 1990). The best known and most fully studied bacteriocin produced by LAB is nisin A (Hurst 1981), which has been accepted by the World Health Organization (WHO) as a preservative in the food industry. Four major classes of bacteriocins are produced by LAB: I, lantibiotics; II, small heat-stable peptides; III, large heat-labile proteins; and IV, complex proteins that additionally require carbohydrate or lipid moieties for bacteriocin activity (Klaenhammer 1993). Most of the bacteriocins of *Lactobacillus* species belong to the class II bacteriocins, which are small, heat-stable, non-lanthionine-containing, membrane-active peptides, and are synthesized as precursors (Jack *et al*. 1995). Unlike typical bacteriocins with a narrow antibacterial spectrum, some LAB bacteriocins exhibit a wider spectrum (Klaenhammer 1993; Jack *et al*. 1995). The broad-spectrum LAB bacteriocins offer great potential as food preservatives, as many of them can inhibit the growth of food-contaminating bacteria (Nettles and Barefoot 1993). Moreover, the probiotic characteristics of lactobacilli from the intestinal tract are very desirable. In particular, *Lactobacillus acidophilus* and related lactobacilli species, such as *Lact. gasseri* and *Lact. johnsonni*, are believed to have colonization and complete competitive abilities in the intestinal tract. Many of them also produce

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