Isolation and Characterization of Bacteriocin Produced by Lactic Acid Bacteria from Fish Gastrointestinal Tract

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ABSTRACT

Screening and characterization of bacteriocin from lactic acid bacteria (LAB) in fish gastrointestinal tract was conducted. Four hundred isolates of LAB were screened from gastrointestinal tract of 6 fish species; Lates calcarifer (seabass), Channa stiata (striped snakehead fish), Clarias batrachus (catfish), Oreochromis niloticus (nile tilapia), Barbonymus gonionotus (javanese barb) and Pangasianodon hypophthalmus (striped catfish). One effective isolate from seabass was able to produce bacteriocin and designated Sb2. Bacteriocin of isolate Sb2 inhibited both gram positive and gram negative bacteria such as Lactobacillus plantarum ATCC 14917, Lb. sakei subsp. sakei JCM 1157^T, Lb. sakei TISTR 890, Lactococcus lactis subsp. cremoris TISTR 1344, Leuconostoc mesenteroides subsp. mesenteroides JCM 6124^T, Leu. mesenteroides TISTR 942, Bacillus coagulans JCM 2257^T, Listeria innocua ATCC 33090, Brochothrix campestris NBRC 11547, Pseudomonas fluorescens JCM 5693^T, Ps. fluorescens TISTR 358, Enterococcus faecalis JCM 5803^T, E. faecalis TISTR 888, Staphylococcus aureus TISTR 118 and Streptococcus sp. TISTR 1030. The maximum bacteriocin activity was obtained at the optimum cultivation condition of 30°C for 18 hr (12,800 AU/ml). Base on morphological, biochemical characteristics (API 50 CHL Kit) and 16s rDNA, the strain Sb2 was identified as Lactococcus lactis spp. lactis, and therefore designated Le. lactis spp. lactis Sb2. The bacteriocin activity was partially inhibited by proteolytic enzyme of trypsin, α-chymotrypsin and proteinase K. It was stable at high temperature up to 100°C for 30 min and at 4°C for 10 days. Probiotic properties of Le. lactis spp. lactis Sb2 were investigated. It was found that Le. lactis spp. lactis
Sb2 was able to grow and produce bacteriocin in pH range of 2-10, concentration of bile salts at 0.3, 0.6 and 0.9% and concentration of NaCl at 1-5%. In addition, antibiotic resistance of isolate Sb2 was determined. The results implied that, *Lc. lactis* spp. *lactis* Sb2 was able to grow and produce bacteriocin when culture in pH 2-10, NaCl 1-5% and 0.3% bile salts. The maximum bacteriocin production (12,800 AU/ml) was observed at pH 6 and in 1% NaCl (12,800 AU/ml). In addition, *Lc. lactis* spp. *lactis* Sb2 was resistant to gentamycin, kanamycin, nalidixic acid, neomycin, norfloxacin, oxolinic acid and sulfamethoxazole/trimethoprim. Sequencing of this gene showed identical sequences to nisin A.