

PROBIOTIC PROPERTIES OF Lactobacillus mucosae STRAINS ISOLATED FROM BRAZILIAN GOAT MILK

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Firstly characterized in 2000, Lactobacillus mucosae is a species presenting properties related to probiotic potential, such as the ability to adhere to mucosal surfaces and activity against pathogenic bacteria. Probiotic characteristics of three L. mucosae strains isolated from goat milk produced in Brazilian semiarid area were investigated. Resistance to simulated gastrointestinal tract (GIT) conditions, bile salt hydrolase (BSH) activity, and β-galactosidase activity of L. mucosae CNPC006, CNPC007, and CNPC009 were studied in vitro. To evaluate the strains tolerance to GIT, overnight cultures of each strain in MRS broth were centrifuged and the pellet was ressuspended in sodium chloride solution (0.85% w/v). Bacterial suspensions was added to simulated gastric juice containing pepsin (3.0 g/l) and lipase (0.9 mg/l), and the pH was adjusted to 2.3 with hydrochloric acid. After 2h of incubation at 37 $^{\circ}$ C with agitation (150 rpm), the enteric conditions were simulated with an artificial intestinal fluid containing pancreatin (1.0 g/l) and bile salts (10.0 g/l) and adjusted to pH 5.0 to simulate the upper intestinal conditions, and to pH 6.5 in the last 2 hours of incubation as above. Aliquots were taken for the enumeration of viable cells at 0, 2, 4, and 6 h. The ability to perform bile salts deconjugation was screened by streaking 10 µl of overnight cultures on MRS agar plates containing either 0.5% (w/v) of the sodium salts of taurocholic acid (TC), taurodeoxycholic acid (TDC), glycocholic acid (GC), or glycodeoxycholic acid (GDC). After anaerobic incubation at 37 $^{\circ}$ C for 72 h, the presence of precipitated bile acid around colonies (opaque halo) was considered a positive result. β-galactosidase activity were assessed qualitatively employing sterile filter paper disks impregnated with o-nitrophenyl-D-galactopyranose. Overnight cultures were streaked on MRS agar plates, and incubated anaerobically at 37 $^{\circ}$ C for 48 h. A colony of each strain was emulsified with 0.1 ml of sterile 0.85% (w/v) sodium chloride solution in a tube containing an ONPG disk. The tubes were incubated at 35° C, and observed for up to 6 hours. The release of a yellow chromogenic compound, o-nitrophenol, indicates a positive result. All the tests was performed two times for each strain, in duplicates. The three L. mucosae strains showed good survival rates when exposed to the conditions simulating the GIT, higher than 50%. The cumulative survival rates obtained for strains CNPC006, CNPC007, and CNPC009 after 6 h were 58.25, 51.03 and 62.83 %, respectively. All the three *L. mucosae* strains showed β-galactosidase activity, were able to grow in presence of TDCA, TCA, GDCA or GCA, and to deconjugate GDC. L. mucosae CNPC007 was able to deconjugate all the tested bile salts. According to the results, the three L. mucosae strains are promising candidates to be further studied as probiotics due to their tolerance to GIT conditions, and BSH and β-galactosidase activity, as well as for the development of novel dairy products. Acknowledgment: EMBRAPA and FAPESP for financial support.