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Physiological and biochemical characterization of the two α -L-rhamnosidases of *Lactobacillus plantarum* NCC245

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ABSTRACT

This work is the first report on the physiological and biochemical characterization of α -L-rhamnosidases in lactic acid bacteria. A total of 216 strains representing 37 species and 8 genera of food grade bacteria were screened for α -L-rhamnosidase activity. *Lactobacillus plantarum* contained the majority of positive strains (25 out of 33) and activity of *L. plantarum* NCC245 strain was examined in more detail. The analysis of α -L-rhamnosidase activity under different growth conditions revealed dual regulation of the enzyme activity, involving carbon catabolite repression and induction: the enzyme activity was down-regulated by glucose and up-regulated by L-rhamnose. The expression of the two α -L-rhamnosidase genes *rhaB1* and *rhaB2* and two predicted permease genes *rhaP1* and *rhaP2*, identified in a probable operon *rhaP2B2P1B1*, was repressed by glucose and induced by L-rhamnose showing regulation at the transcriptional level. The two α -L-rhamnosidase genes were overexpressed and purified from *E. coli*. *RhaB1* activity was maximal at 50 °C and at neutral pH and *RhaB2* maximal activity was detected at 60 °C and at pH of 5, showing high residual activity at 70 °C. Both enzymes showed preference for the α -1,6 linkage of L-rhamnose to β -D-glucose, hesperidin and rutin being their best natural substrates, but surprisingly no activity was detected towards the α -1,2 linkage in naringin. In conclusion, we identified and characterized the strain *L. plantarum* NCC245 and its two α -L-rhamnosidase enzymes, which might be applied for improvement of bioavailability of health beneficial polyphenols, such as hesperidin in humans.

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