Improvement of the lytic properties of a ?-1,3-glucanase by directed evolution

Salazar, Oriana

Basso, Caterina

Barba, Paola

Orellana, Claudia

Asenjo, Juan A.

BGLII is a bacterial endoglucanase that hydrolyzes the ?-1,3-glucan present in yeast cell walls, resulting in lysis of Saccharomyces cerevisiae. As a result of this property, BGLII is considered a potential tool for downstream processing and recovery of biotechnological products produced in yeast. Here we describe the improvement of the yeast lytic activity of BGLII, achieved by a directed evolution approach involving random mutagenesis and screening for variants with improved catalytic activity, combined with site-directed mutagenesis. A BGLII variant having three times the wild-type hydrolytic activity on laminarin was identified. The purified enzyme also exhibited higher lytic activity on yeast cells. Mutations causing the improvements are located very close to each other in the amino acid sequence, suggesting that the region should be considered as a target for further improvements of the glucanase activity. These results demonstrate the feasibility of molecular evolution methods f