P4.7 - EVALUATION OF YEAST SURFACE DISPLAY OF THE ENDOLYSIN PLY511 AGAINST *LISTERIA MONOCYTOGENES*

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ABSTRACT

Yeast surface display, also known as yeast display, is a protein engineering technique that uses the expression of recombinant proteins of interest, fused with native cell wall proteins of yeast. Ply511 endolysin is a cell wall hydrolase with N-acetylmuramoyl-L-alanine amidase activity.

Ply511 is naturally produced by the Listeria phage A511 to lyse bacterial cells and has been proposed as an alternative to antibiotics.

Listeria monocytogenes is a Gram-positive pathogen causative of human infections, resulting in febrile gastroenteritis, perinatal infection, and central nervous system infections. These infections are frequently acquired through the ingestion of contaminated food.

In our study, we employed CRISPR-Cas9 to genetically modify Saccharomyces cerevisiae to display the listeria endolysin Ply511 on its surface. Flow cytometer analyses confirmed the expression of the genetic construction integrated in the recombinant yeast. Also, the enzymatic peptidoglycan-degrading activity of the engineered yeast was confirmed by using heat-killed L. monocytogenes cells. In killing assays, the recombinant yeast did not show CFU reduction of Listeria in comparison with the wild-type. These results suggest that yeast display of endolysins needs to be improved to be used as an antimicrobial strategy in the context of engineered probiotics, to assure that bacterial killing is achieved.

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