Functional characterization of a *Yarrowia lipolytica* gene family coding for carboxylic acids permeases homologues

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Carboxylic acids are widely used in pharmaceutical and food industries and new industrial uses are continually emerging. The first step of carboxylic acids metabolism is their entrance to the cell and understanding in detail this process has significant biotechnological relevance. The first carboxylic acids permease found in Saccharomyces cerevisiae was a lactate/H+ symporter coded by JEN1 gene. In Kluyveromyces lactis, as well as in Candida albicans, two genes were described that encode two JEN1-like transporters specific for mono-e and dicarboxylate uptake, KlJEN1/KlJEN2, and CaJEN1/CaJEN2 respectively^{1,2,3}. The sequencing of other hemiascomycetes genomes by the Genolevures project has demonstrated the existence of a family of Jen1p homologs in other different yeasts. Some of these yeasts have more than two homologues, but its function is still unclear. This is the case of Yarrowia lipolytica, a yeast species able to use a broad range of substrates and that presents 6 homologues to Jen1p. A mediated transport system for mono-, di- and tricarboxylates was found in this yeast, suggesting that the respective gene family can putatively code for carboxylate permeases. In order to assess their functional characterization, these genes have been heterologously expressed in the strain S. *cerevisiae jen1\Delta ady2\Delta* that presents no activity for a carboxylate permease⁴. All the transformant strains showed a slightly improved growth in mono- and di- but not tricarboxylic acids containing media. Alongside, RT-PCR assays showed that YLJEN expression is induced by carboxylic acids and YlJen-GFP chimeric proteins were all located in the plasma membrane. Taken together, these results suggest that these Jen1 homologues can mediate the transport of carboxylic acids in Y. lipolytica.

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