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Functional characterization of Cyberlindnera jadinii carboxylate transporters in Saccharomyces cerevisiae

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Introduction

In Saccharomyces cerevisiae, two permeases are responsible for the uptake of carboxylates (CA) at the plasma membrane, Jen1p a monocarboxylate proton symporter (Major Facilitator Superfamily) and Ady2p an acetate permease (AceTr Family).

In *Cyberlindnera jadinii*, different uptake systems for CAs were functionally characterized however until now the genes encoding these transporters remain unidentified. In this work, CA transporter homolog genes from *C. jadinii* were identified and expressed in *S. cerevisiae*.

Materials and Methods

The *S. cerevisiae* strain W303-1A $jen1\Delta$ $ady2\Delta$, lacking carboxylate uptake capacity, was used to express *C. jadinii ScJEN1* and *ScADY2* homologs. Genes were identified through sequence alignment and homology prediction and cloned in the p416GPD vector, under the control of a GPD constitutive promoter. GFP-fusions versions were used to determine protein expression and localization. Transport activity was determined through growth on different carbon sources and measurement of the uptake of labelled CAs, namely D,L-[U-14C] lactic acid, [2,3-14C] succinic acid and [1-14C] acetic acid.

Results

In *C. jadinii*, 6 genes homolog to *ScJEN1* (Cjj23088, Cjj21966, Cjj22358, Cjj21989, Cjj21602, Cjj25129) and 4 genes homolog to *ScADY2* (Cja24587, Cja20823, Cja20690, Cja20822) were identified. All proteins were expressed and localized at the plasma membrane. Regarding transporter specificity *CJJEN1*-6 and *CJAD3* encode lactate transporters, *CJAD1* and 4, lactate and acetate transporters, and *CJAD2* encodes a lactate, acetate and succinate transporter.

Conclusions

In this work, we identified 6 CjJEN1 and 4 CjADY2 homologs that are functional carboxylate transporters in S. cerevisiae. All the CjJEN1 homologs are lactate transporters and CjADY2 homologs

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Portuguese Society of Genetics

present different specificities. Further studies are underway to fully characterize these ten new plasma membrane transporters from *C. jadinii*.

References

Soares-Silva, I., et al. (2007). Mol Membr Biol. 24(5-6), 464-474.