

Regulation of gene expression by sugarcane juice in *Gluconacetobacter diazotrophicus*

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Gluconacetobacter diazotrophicus is an endophytic, nitrogen-fixing bacterium that lives in association with sugarcane, sweet potato, elephant grass, coffee and pineapple. Strain PAL5 (ATCC 49037) was isolated from roots of sugarcane grown in Brazil, and has great potential to be used as a biofertilizer, as it fixes nitrogen and promotes plant growth mainly under low N levels in soils. Its genome DNA sequence has been publicly available, and now our interests focus on regulation of its gene expression at genomic level. Recently, we described the construction of a *G. diazotrophicus* genome library in the promoter-trap vector pPW452, which bears the promoter-less *lacZ* reporter gene; the whole library has been transferred into *G. diazotrophicus* PAL5 cells. By using this promoter library, we have screened for differential gene expression in *G. diazotrophicus*, monitored through β -galactosidase activity, in the presence of juice from different sugarcane genotypes. Here we show preliminary results of this experiment. In brief, near 1,100 library clones were grown in the presence of filter-sterile juice from either a wild variety or a commercial cultivar of sugarcane, comparing with growth in 10 mM glucose, and assayed for β -galactosidase activity using 5-bromo-4-chloro-3-indolyl- β -D-galactopyranoside (X-GAL, in solid medium) and/or *o*-nitrophenyl- β -D-galactopyranoside (ONPG, after growth in liquid medium) as substrates. So far, a clone presented activation of *lacZ* gene expression levels in the presence of juice from either sugarcane varieties tested, while further assays involving other four clones are being performed in order to confirm their differential gene expression levels under the growth conditions tested. These clones will be subjected to further analysis in order to identify their promoter regions through DNA sequencing.

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