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PO02

Poster

Impala*-based transposon mutagenesis is influenced by chromatin modifications in the fungal plant pathogen *Zymoseptoria tritici

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Transposition of *TC1-mariner impala* from *F. oxysporum* was tested in *Zymoseptoria tritici*. We used an existing excision assay in which *impala* is inserted in *A. nidulans* nitrate reductase gene promoter. This vector was introduced in a *Z. tritici* nitrate reductase mutant unable to grow on nitrate minimal medium (MM). Inoculating these transgenic strains onto MM allowed recovering revertants (10-30/plate). Almost all revertants corresponded to excisions of *impala* from *niaD* (95%). Inoculum culture conditions had a significant effect on *impala* excision rate, the optimum being obtained with yeast-like cells grown on YPD for 5 days at 18°C. Varying culture conditions during reversion assay (carbon starvation, light, darkness, heat or cold shocks, and copper stress) had no effect on *impala* excision rate. However, adding histone deacetylase inhibitor trichostatin at a sub-inhibitory concentration (0.1 µM) to MM during reversion assays, significantly increased *impala* excision rate. This unexpected result suggests that modifying histone acetylation level has an effect on *impala* excision either as a consequence of modifying chromatin status at vector integration site, or through a direct effect on *impala* machinery. In most revertants (90%), *impala* was inserted in *Z. tritici* genome. 60 *impala* re-insertion loci were characterized by Ligation-mediated PCR amplification and sequencing. *Impala* was inserted at random locations in core chromosomes (no hot spots), but not in accessory chromosomes (0/8 expected insertions). Since accessory chromosomes mostly carry repressed chromatin, this result suggests that *impala* preferentially inserts in regions with open chromatin. *Impala* also inserts at lower rate in native transposons (7%) compared to random (30%), suggesting that it avoids these regions. Overall, *impala* was preferentially inserted either at the 5' end of transcriptionally active genes (5'UTR, promoters; 68%) or in genes (14%). These results show that *impala* is active in *Z. tritici*. Its integration patterns make it particularly suitable for insertional mutagenesis.