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POSTER

REGULATION OF THE AUROFUSARIN GENE CLUSTER IN FUSARIUM GRAMINEARUM

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<u>Fusarium graminearum</u> and related species produces the red pigment <u>aurofusarin</u>. Aurofusarin is a homodimeric polyketid. Its synthesis is dependent on *pks12* [1,2], *aurJ*, *fmo* and *gip1* [1], all located in the 24 kb gene cluster [1].

The biosynthesis of the polyketide aurofusarin is regulated by multiple factors. A bioinformatic analysis of the aurofusarin gene cluster, has revealed two transcription factors (aurR1 and aurR2) of the binuclear zinc cluster type (Cys_6Zn_2) [1]. Agrobacterium mediated transformation was used to replace these genes with hygR. The mutants have been analysed for morphological changes, by HPLC-UV and for expression of genes in the aurofusarin gene cluster by RT-PCR.

Replacement of *aurR1* results in a milky white mutant and aurofusarin is not detectable by HPLC. RT-PCR analysis shows that the lack of aurofusarin production is due to a lack of expression of *pks12*, *aurJ*, *fmo* and *gip1*. Replacement of *aurR2* results in a red mutant, with a growth rate similar to the wild type. HPLC analysis shows that the mutant produces less aurofusarin than the wild type, but RT-PCR analysis does not show any changes in expression of genes in the aurofusarin gene cluster.

Random mutagenesis has resulted in the identification of two additional genes (pr5.1 and pr35.1), located outside the aurofusarin gene cluster, being required for production of aurofusarin. The pr5.1 mutant is mutated in a single gene which is found in a number of pathogenic fungi. The strain has a white phenotype and does not produce aurofusarin, due to a lack of expression of PKS12 and additional PKS-genes. The pr35.1 mutant produces a novel yellow pigment. The lack of aurofusarin production is however not due to a lack of expression from the gene cluster. This indicates that pr35.1 is involved in regulation on a post-transcriptional or directly in the biosynthesis.

^[1] Malz et al. 2005. Identification of a gene cluster responsible for the biosynthesis of aurofusarin in the *Fusarium graminearum* species complex. Fungal Genet. Biol. 42(5):420-433

^[2] Kim *et al.* 2005. Putative polyketide synthase and laccase genes for biosynthesis of aurofusarin in *Gibberella zeae*. Appl. Environ. Microbiol. 71(4):1701-1708