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IMPACT OF CHROMOSOMAL POSITION EFFECT ON HETEROLOGOUS PRODUCTION IN *S. ALBUS* J1074

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Streptomyces albus J1074 is a typical representative of *Streptomyces* genus and is well known for its outstanding potential for heterologous production. For the last decade it was used for heterologous expression of numerous antibiotic biosynthetic clusters, *e. g.*, thiocoraline, cyclooctatin and steffimycin. Numerous genetic tools recently developed and perfect heterologous host properties of the strain make it potentially to a good candidate as universal heterologous host. However, employment of this potential requires optimization of antibiotic production. One of the factors that can influence expression of heterologous genes is chromosomal position effect. This term describes differences in gene expression caused by location of the genes on the chromosome. It may affect expression of native genes as well as transgenes inserted into different regions of genome. Despite their huge importance as natural products producers, theposition effect was not yet investigated in streptomycetes due to lack of effective tools for genetic manipulations.Recent accumulation of genome sequencing data and adaptation of transposon mutagenesis system for streptomycetes represent the opportunity to explore this phenomenon closely and to obtain the strain with enhanced antibiotic production.

With this aim the β-glucuronidase reporter gene was placed on minimariner transposon between two *fd*-phageterminators and transposon mutant library with randomly distributed reporter gene was obtained. The GusA-activity of 25 mutants was measured and 6-fold variation of activity had been observed. Obtained results were also checked for correlation with factors that may impact position effect phenomenon (distance to *oriC*and strength of local promoters).

To obtain the strain with improved antibiotic production the *attB*site of φC31 was introduced into minimariner transposon and transposon mutant library with randomly distributed *attB*-containing transposons was generated. After integration of the cosmid, containing aranciamycin biosynthetic cluster, into genomes of obtained mutants *viaattB*x*attP* recombination, production levels of this antibiotic in 26 mutantswere compared. It was observed that relocation of aranciamycin biosynthetic cluster through over the chromosome led mostly to significant decrease of aranciamycin production. Meanwhile, strains with enhanced number of *attB*-sites demonstrated higher antibiotic production.

As a result, the impact of chromosomal position on expression of heterologous genes had been explored and factors that may cause this effect were analyzed.Using combination of system for transposon mutagenesis and *attB*×*attP* recombination system aranciamycin biosynthetic cluster was introduced randomly into the *S. albus*genome. Analysis of aranciamycin producers demonstrated that copy number of antibiotic biosynthetic cluster is crucial for increase of antibiotic production.

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