

Novel auto-induction expression systems with high expression strength and wide induction initiation in *Bacillus subtilis*

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Abstract

The low expression strength and fixed induction initiation have always been the main obstacle for applying the bacterial quorum sensing (QS) system to protein production. As a typical QS system in *Bacillus subtilis*, ComQXPA activates the promoter PsrfA using ComX and ComA as the auto-inducer and promoter activator, respectively. This study developed a series of novel auto-induction expression systems in *B. subtilis* WB600 based on ComQXPA using super-folder green fluorescent protein as the reporter. First, the -35 region of PsrfA was replaced by the corresponding conserved sequence of σ A-dependent promoters, yielding P1 with an 85% enhanced expression strength. Second, by conducting a semi-rational design within the spacer between -35 and -15 regions of in P1, we generated the ComQXPA promoter PS1E, the expression strength of which is 8.22-fold higher than that of PsrfA. Based on PS1E, we finally obtained three types of auto-induction expression systems with the induction initiations ranging from 1.5 h to 9.5 h by optimizing the combination of the promoters for ComX and ComA. By using the auto-induction expression systems, the yield of *Bacillus deramificans* pullulanase in *B. subtilis* reached 80.2 U/mL, 0.36-fold higher than the strongest constitutive promoter P566. With the diversity in dynamic features, the novel auto-induction expression systems provide great potential for improving protein expression and metabolite production in *B. subtilis*.

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