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Construction of a plasmid interspecific transfer system in *Bacillus* species with the counter-selectable marker mazF

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Bacillus species strains as attractive hosts to produce heterologous secretory proteins usually play important roles in bio-industry. However, low transformation efficiency of exogenous plasmids limited the application of *Bacillus* species. Here, a novel plasmid interspecific transfer system, with higher transformation efficiency, higher positive rate and more convenient manipulation has been successfully constructed. A high transformation efficiency strain *Bacillus subtilis* F-168 containing the counter-selectable marker mazF was used as the plasmid donor strain in this transfer method. A shuttled plasmid pBE980b and its recombinant plasmids pBE980b::BapuI and pBE980::Spro1 were successfully transferred into the recipient *Bacillus strains* (*Bacillus amyloliquefaciens* 66, *Bacillus licheniformis* 124 and *Bacillus megaterium* 258) by this method. After co-culturing the donor cells (OD 600 nm=1.4-1.7) and the recipient cells (OD 600 nm=0.5-0.9) cells for 24 hours in 22°C, more than 1.0×10^5 positive transformants were obtained and a transformation efficiency of 1.0×10^{-3} showed 10-100 times higher than those transferred using traditional transfer methods. This plasmid interspecific transfer system would provide a new approach for genetic manipulation in *Bacillus* strains and accelerate the research progress of *Bacillus* strains in bio-industry.

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