# Strain construction

The plasmids and strains used in this study are described in Table 1 below. Plasmid isolations, enzymatic manipulations of DNA, agarose gel electrophoresis and other routine DNA manipulations were performed according to the methods of Sambrook and Russell [[1](#_ENREF_1)]. The QIAquick Gel Extraction Kit and QIAquick PCR purification kit (Qiagen) were used for DNA-purifications from agarose gels and enzymatic reactions, respectively. PCR for cloning and allele identification was performed using the Expand High Fidelity PCR-system (Boehringer Mannheim). DNA was sequenced using the Big-Dye Terminator v1.1 Cycle kit (Applied Biosystems). Transformations of *E. coli* were performed using the rubidium-chloride method (available at [www.neb.com](http://micro.nwfsc.noaa.gov/protocols/rbcl.html)). Matings and selection of double recombinants were performed as described earlier [[2](#_ENREF_2), [3](#_ENREF_3)].

Table 1. Strains and plasmids in this study.

|  |  |  |
| --- | --- | --- |
| **Strains** | **Description** | **Ref** |
| *E. coli* S17.1 | RP4 2-Tc::Mu-Km::*Tn7* *pro res mod+* | [[4](#_ENREF_4)] |
| *P. fluorescens* SBW25 | Wild type | [[5](#_ENREF_5)] |
| *P. fluorescens* SBW25*mucA* | Derivative of wild type strain using pAT71 to introduce a stop codon in *mucA.* | This work |
| *P. fluorescens* SBW25 Δ*algC mucA* | Derivative of  *P. fluorescens* SBW25*mucA* where pKB22 were used to delete parts of *algC.* | This work |
| *P. fluorescens* SBW25Δ*algC* | Derivative of  *P. fluorescens* SBW25 Δ*algC mucA* using pAT70 to repair *mucA.* | This work |
| *P. fluorescens* SBW25*mucA* AlgD- | Derivative of  *P. fluorescens* SBW25*mucA* using pMBN15 to insert a transcription terminator between PalgD and *algD.* | This work |
| **Plasmids** |  |  |
| pHE179 | ColEI cloning vector. Tcr, Apr | [[6](#_ENREF_6)] |
| pMG48 | RK2-based suicide vector encoding β-galactosidase; Tcr  Apr | [[3](#_ENREF_3)] |
| pAT70 | pMG48 based vector used to repair *mucA*. Tcr  Apr | [[6](#_ENREF_6)] |
| pAT71 | pMG48 based vector used to introduce a stop codon in *mucA*. Tcr  Apr | [[6](#_ENREF_6)] |
| pKB22 | pMG48 based vector used to delete *algC* | [[2](#_ENREF_2)] |
| pHE139 | RK2 derivative encoding P*algD* from *P. fluorescens* | [[6](#_ENREF_6)] |
| pHE142 | RK2 derivative encoding *algD* from *P. fluorescens* | [[6](#_ENREF_6)] |
| pMBN14 | Derivative of pHE179 where a 0.8 kb DNA fragment containing the *algD*-promoter, and a 1.6 kb DNA-fragment encoding *rrnB* and *algD* were inserted. | This work |
| pMBN15 | Derivative of pMG48 in which a 2.4 kb DNA fragment from pMBN14 containing *PalgD-rrnB-algD* was inserted. Used to create a strain with no promoter before *algD*. | This work |

# Acknowledgments:

Heidi Myrset participated in making the plasmids and strains used in this study.

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