

Table S3 Plasmids

| Plasmid | Relevant features                                                                        | Parent vector | Restriction enzymes to digest parent plasmid | Primers used   | PCR template | Assembly | Reference |
|---------|------------------------------------------------------------------------------------------|---------------|----------------------------------------------|----------------|--------------|----------|-----------|
| pAP114  | $P_{xyl}::mCherryOpt\ cat$                                                               |               |                                              |                |              |          | 50        |
| pCE971  | $P_{xyl}::ugtB\ cat$                                                                     | pAP114        | BamHI, SacI                                  | CDEP5693-5694  | R20291       | ITA      |           |
| pBZ139  | $P_{xyl}::ugtA\ cat$                                                                     | pAP114        | BamHI, SacI                                  | CDEP6171-6172  | R20291       | ITA      |           |
| pCE678  | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-pgdA-2$ homology to delete <i>pgdA</i> <i>cat</i>     |               |                                              |                |              |          | 52        |
| pCE1062 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-pgdA-2$ homology to delete <i>ugtB</i> <i>cat</i>     | pCE678        | NotI, XhoI                                   | CDEP6257-6260  | R20291       | ITA      |           |
| pCE1065 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-ugtB\ cat$                                            | pCE1062       | MscI, MluI                                   | CDEP4237, 6271 | pCE678       | ITA      |           |
| pCE1069 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-pgdA-2$ homology to delete <i>ugtA</i> <i>cat</i>     | pCE678        | NotI, XhoI                                   | CDEP6290-6293  | R20291       | ITA      |           |
| pCE1071 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-ugtA\ cat$                                            | pCE1069       | MscI, MluI                                   | CDEP4237, 6274 | pCE678       | ITA      |           |
| pCE1088 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-pgdA-2$ homology to delete <i>cdr_0773</i> <i>cat</i> | pCE678        | NotI, XhoI                                   | CDEP6458-6461  | R20291       | ITA      |           |
| pCE1098 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-cdr_0773\ cat$                                        | pCE1088       | MscI, MluI                                   | CDEP4237, 6462 | pCE678       | ITA      |           |
| pCE1085 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-pgdA-2$ homology to delete <i>cdr_2958</i> <i>cat</i> | pCE678        | NotI, XhoI                                   | CDEP6454-6457  | R20291       | ITA      |           |
| pCE1105 | $P_{xyl}::Cas9-opt\ P_{gdh}::sgRNA-cdr_2958\ cat$                                        | pCE1085       | MscI, MluI                                   | CDEP4237, 6469 | pCE678       | ITA      |           |
| pDR111  | <i>amyE</i> :: $P_{IPTG}$ <i>amp spec</i>                                                |               |                                              |                |              |          |           |
| pCE1057 | <i>amyE</i> :: $P_{IPTG}::ugtB\ amp\ spec$                                               | pDR111        | HindIII, SphI                                | CDEP6229-6230  | R20291       | ITA      |           |
| pCE1059 | <i>amyE</i> :: $P_{IPTG}::ugtA\ amp\ spec$                                               | pDR111        | HindIII, SphI                                | CDEP6227-6228  | R20291       | ITA      |           |
| pAC68   | <i>thrC</i> :: $P_{xyl}\ amp\ erm$                                                       |               |                                              |                |              |          |           |
| pCE1122 | <i>thrC</i> :: $P_{xyl}::ugtB\ amp\ erm$                                                 | pAC68         | HindIII, BamHI                               | CDEP6615-6616  | R20291       | ITA      |           |
| pCE1281 | <i>thrC</i> :: $P_{xyl}::ugtA\ amp\ erm$                                                 | pAC68         | HindIII, BamHI                               | CDEP6628-6629  | R20291       | ITA      |           |
| pCE1062 | <i>thrC</i> :: $P_{xyl}::hexSDF\ amp\ erm$                                               | pAC68         | HindIII, BamHI                               | CDEP6215-6216  | R20291       | ITA      |           |