**Plasmids and strains deposited by the Hegemann lab**

Visit [www.chlamy.de](http://www.chlamy.de) for more info or contact CRISPR@chlamy.de

**pHS-SpCas9 (pPH185)** {link:https://www.chlamycollection.org/product/phs-spcas9/}

Expression of *Streptococcus pyogenes* (Sp)Cas9 optimized to *C. reinhardtii* codon bias, controlled by the HSP70A heat-shock promoter. *map* {link:<https://benchling.com/s/V6Yn3QYk>}

**pHR-SpCas9 (pPH186)** {link: https://www.chlamycollection.org/product/phr-spcas9/}

Expression of *Streptococcus pyogenes* (Sp)Cas9 optimized to *C. reinhardtii* codon bias, controlled by the HSP70A/RBCS2 tandem promoter. *map* {link:<https://benchling.com/s/3XkMrsaP>}

**pHS-SaCas9 (pPH187)** {link: }

Expression of *Staphylococcus aureus* (Sa)Cas9 optimized to *C. reinhardtii* codon bias, controlled by the HSP70A heat-shock promoter. *map* {link:[https://benchling.com/s/qNMlZpm8}](https://benchling.com/s/qNMlZpm8%7D)

**pHR-SaCas9 (pPH188)** {link: }

Expression of *Staphylococcus aureus* (Sa)Cas9 optimized to *C. reinhardtii* codon bias, controlled by the HSP70A/RBCS2 tandem promoter. *map* {link:[https://benchling.com/s/btotv3OR}](https://benchling.com/s/btotv3OR%7D)

**pCrU6.1-SpCas9 cloning (pPH195)** {link: }

Cloning vector for *Streptococcus pyogenes* Cas9 (SpCas9) single guide RNA transcription, controlled by the U6snRNA promoter #1. *map* {link:<https://benchling.com/s/aw1TqJHw>}

**pCrU6.2-SpCas9 cloning (pPH158)** {link: }

Cloning vector for *Streptococcus pyogenes* Cas9 (SpCas9) single guide RNA transcription, controlled by the U6 snRNA promoter #2. *map* {link: <https://benchling.com/s/gB07y8zl>}

**pCrU6.3-SpCas9 cloning (pPH159)** {link: }

Cloning vector for *Streptococcus pyogenes* Cas9 (SpCas9) single guide RNA transcription, controlled by the U6 snRNA promoter #3. *map* {link: [https://benchling.com/s/elcQsHBB}](https://benchling.com/s/elcQsHBB%7D)

**pCrU6.4-SpCas9 cloning (pPH160)** {link: }

Cloning vector for *Streptococcus pyogenes* Cas9 (SpCas9) single guide RNA transcription, controlled by the U6 snRNA promoter #4. *map* {link: [https://benchling.com/s/OPkVaRUz}](https://benchling.com/s/OPkVaRUz%7D)

**pCrU6.4-SpCas9 cloning/aphVIII (pPH340)** {link: }

Cloning vector for *Streptococcus pyogenes* Cas9 (SpCas9) single guide RNA transcription, controlled by the U6 snRNA promoter #4 with *aphVIII* cassette for selection on paromomycin. *map* {link: [https://benchling.com/s/seq-3xKbLvsJMKAEs4O7x8GQ}](https://benchling.com/s/seq-3xKbLvsJMKAEs4O7x8GQ%7D)

**pCrU6.4-SpPSY1/aphVIII (pPH332)** {link: }

Vector for single guide RNA targeting *PSY1* with RNA scaffold for *Streptococcus pyogenes* Cas9 (SpCas9), controlled by the U6 snRNA promoter #4 with *aphVIII* cassette for selection on paromomycin. *map* {link: <https://benchling.com/s/seq-kcWAvleU4pAJAIrKpPuh>}

**pCrU6.1-SaCas9 cloning (pPH193)** {link: }

Cloning vector for *Staphylococcus aureus* Cas9 (SaCas9) guide RNA transcription, controlled by the U6 snRNA promoter #1. *map* {link: [https://benchling.com/s/G0EsVCMb}](https://benchling.com/s/G0EsVCMb%7D)

**pCrU6.2-SaCas9 cloning (pPH154)** {link: }

Cloning vector for *Staphylococcus aureus* Cas9 (SaCas9) guide RNA transcription, controlled by the U6 snRNA promoter #2. *map* {link: [https://benchling.com/s/amFo61xA}](https://benchling.com/s/amFo61xA%7D)

**pCrU6.3-SaCas9 cloning (pPH155)** {link: }

Cloning vector for *Staphylococcus aureus* Cas9 (SaCas9) guide RNA transcription, controlled by the U6 snRNA promoter #3. *map* {link: [https://benchling.com/s/Y4W5dQQh}](https://benchling.com/s/Y4W5dQQh%7D)

**pCrU6.4-SaCas9 cloning (pPH156)** {link: }

Cloning vector for *Staphylococcus aureus* Cas9 (SaCas9) guide RNA transcription, controlled by the U6 snRNA promoter #4. *map* {link: [https://benchling.com/s/K8B7iSVp}](https://benchling.com/s/K8B7iSVp%7D)

**pCrU6.4-SaCas9 cloning/aphVIII (pPH339)** {link: }

Cloning vector for *Staphylococcus aureus* Cas9 (SaCas9) guide RNA transcription, controlled by the U6 snRNA promoter #4 with *aphVIII* cassette for selection on paromomycin. *map* {link: [https://benchling.com/s/seq-tw570ykR6MRUqx0FGeaF}](https://benchling.com/s/seq-tw570ykR6MRUqx0FGeaF%7D)

**pCrU6.4-SaPSY1/aphVIII (pPH331)** {link: }

Vector for guide RNA targeting *PSY1* with scaffold for *Staphylococcus aureus* Cas9 (SaCas9), controlled by the U6 snRNA promoter #4 with *aphVIII* cassette for selection on paromomycin. *map* {link: <https://benchling.com/s/seq-MsLOs4rE4YFL47tnpKhH>}

**pAphVII (pPH360)** {link: }

Codon optimized Streptomyces aminoglycoside phosphotransferase *aphVII* marker gene for Hygromycin B selection. *map* {link: [https://benchling.com/s/seq-hi1yE3a0oq99MoxzxJmi}](https://benchling.com/s/seq-hi1yE3a0oq99MoxzxJmi%7D)

**pAphVIII (pPH075)** {link: }

Codon optimized Streptomyces aminoglycoside-5 ́- phosphotransferase *VIII* antibiotic selection marker gene for paromomycin selection. *map* {link: [https://benchling.com/s/seq-XD7NEQsswVZd6nNurb1w}](https://benchling.com/s/seq-XD7NEQsswVZd6nNurb1w%7D)

**Reference**

Greiner, A., Kelterborn, S., Evers, H., Kreimer, G., Sizova, I., and Hegemann, P.Targeting of photoreceptor genes via zinc-finger nucleases and CRISPR/Cas9 in *Chlamydomonas reinhardtii.* Plant Cell 30 (2017). <https://doi.org/10.1105/tpc.17.00659>

**Overview of all CRISPR/Cas9 plasmids from the Hegemann lab**

<https://www.chlamycollection.org/hegemann_lab>

Visit [www.chlamy.de](http://www.chlamy.de) for more info or contact CRISPR@chlamy.de