



Supplementary Fig 1. Strategy for generation and analysis of strains WT-RCS3 and NbIR^{D57A}-RCS3. (A) The integration of the CS3 cassette (hatched bars) in the intergenic region (solid bar) between *nbIR* and the downstream ORFs Synpcc7942_2306 (open arrows) by homologous recombination is shown schematically. Flanking chromosome regions are represented by dotted lines and plasmid sequences by a continuous line. Depending on specific crossover sites two alternative *nbIR* alleles can be generated. Mutant (*nbIR*^{D57A}) and wild type strains differ at the indicated *PvuI* site. (B) Schematic representation of the allele structure in strains WT-RCS3 and NbIR^{D57A}-RCS3. Relevant *PvuI* sites are shown. Positions of primers used to verify allele structure are indicated by black arrows (C) PCR analysis of WT-RCS3 (lane 1), NbIR^{D57A}-RCS3 (lane 2) and *Synechococcus* sp. PCC7942 (lane3) using primers NbIR-1F (1F) and CS3-2R (2R). (D) *PvuI* digestion of the PCR fragment generated with primers 1F and NbIR-1R (1R). Lane numbers as in (C). M: size marker λ *HindIII*+*EcoRI*. L: DNA 100 bp ladder (Fermentas).