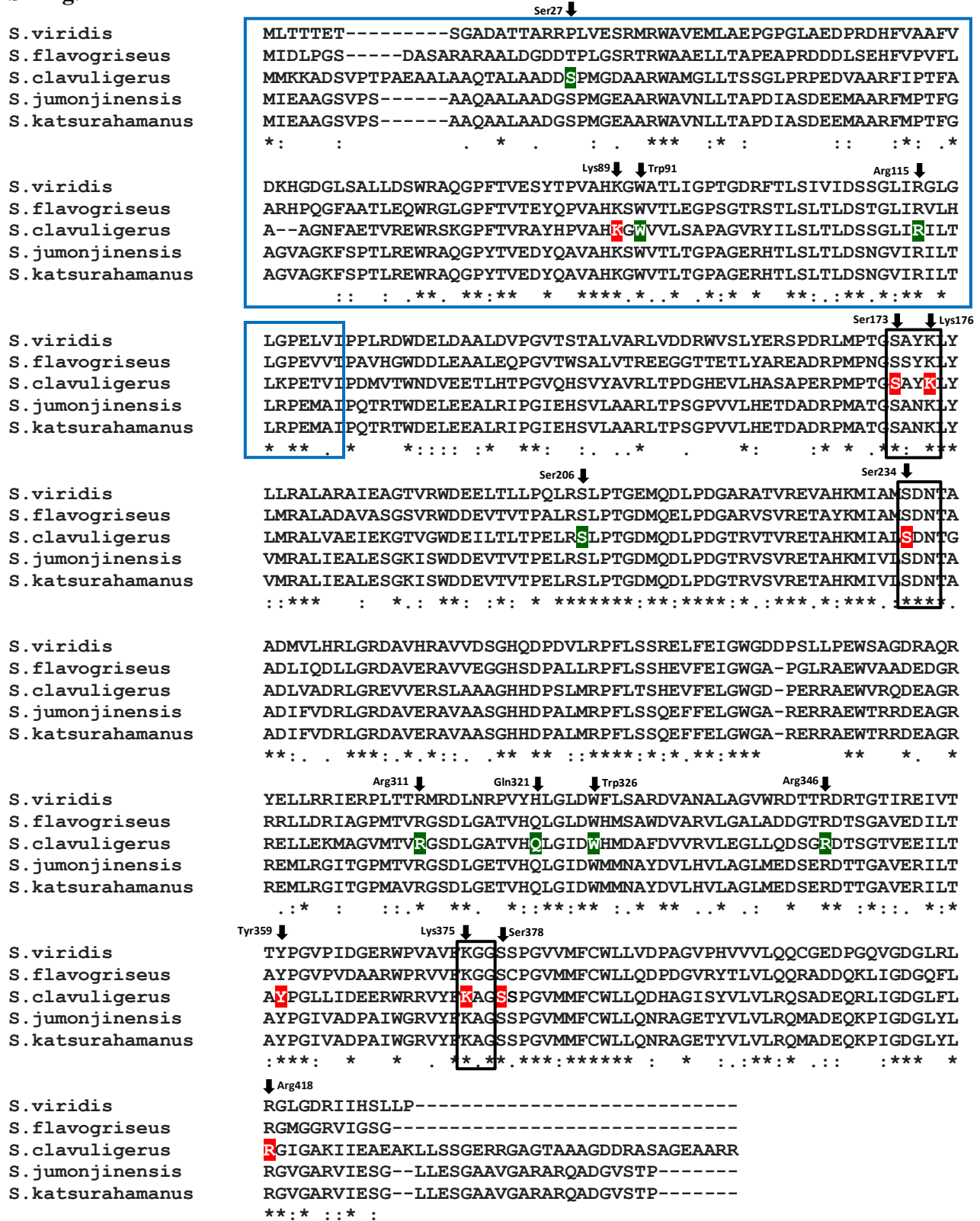


S4 Fig.



**S4 Fig.** Multiple sequence alignment of different Cpe proteins described in the current study. Analysis was performed with Clustal Omega (ver 1.2.1) using translated Cpe amino acid sequences from *S. clavuligerus* (WP\_003952519.1), *S. flavogrisius* (WP\_014152684.1), *S. viridis* (WP\_015787620), *S. jumonjinensis* and *S. katsurahamanus*. The DNA sequences of *cpe* from the latter two producers (*S. jumonjinensis* and *S. katsurahamanus*) were determined as part of the current study and are reported in S3 Fig. The boxes in black highlight the conserved SXXK, SDN and KTG/KAG motifs present in class A  $\beta$ -lactamses and the respective Cpe proteins, whereas the box in blue represents the N-terminus domain indentified in Cpe<sup>Sc</sup>. The arrows indicate amino acids from Cpe<sup>Sc</sup> that were selected for mutagenesis and the ones highlighted in red were shown to be essential for *in vivo* clavulanic acid production in *S. clavuligerus*.