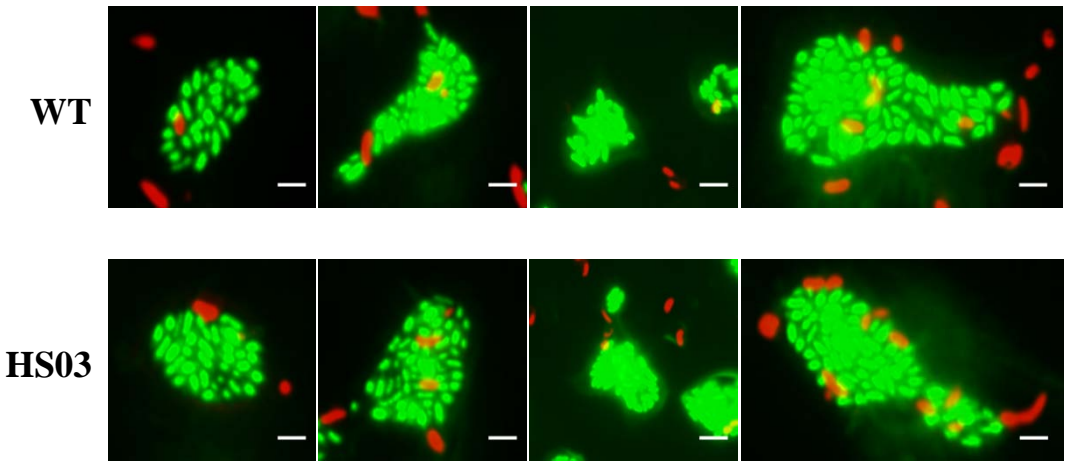


A



B

RNA [$\mu\text{g} / \mu\text{l}$]

0.289

0.305

C

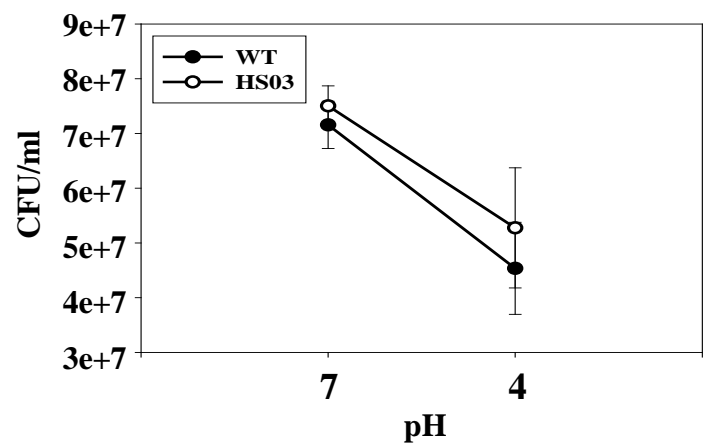


Figure S1. Uniformity of the biofilm inocula used for *in vivo* experiments. (A) Micrographs of biofilm suspension used as inocula for mouse infection experiments. Viability of the inocula was analyzed using a LIVE/DEAD *BacLight* viability kit (Molecular Probes, Eugene, OR) as described elsewhere (1). After staining, randomly selected areas were imaged using an epifluorescence microscope ($\times 1000$ magnification, Olympus BX51, Tokyo, Japan) to differentiate viable (with an intact cell membrane, green) and nonviable (lacking an intact cell membrane, red) cells. (B) Total RNAs were isolated from the inocula using an RNeasy Mini Kit (Qiagen), and the concentrations of the RNA were measured by NanoVue PlusTM Spectrophotometer (GE Healthcare, Buckinghamshire, UK). (C) Acid resistance of biofilms of the wild type and *smcR* mutant. Biofilms preformed as described in Materials and Methods were soaked with VFMG adjusted to pH 7 or 4, respectively. After incubation for 30 min at 30°C, the survival of the biofilm-derived cells was determined by enumeration of CFU. WT, wild type; HS03, *smcR* mutant.

REFERENCE

1. Lim MS, Kim JA, Lim JG, Kim BS, Jeong KC, Lee KH, Choi SH. 2011. Identification and characterization of a novel serine protease, VvpS, that contains two functional domains and is essential for autolysis of *Vibrio vulnificus*. J. Bacteriol. **193**:3722-3732.