## Chirality in microbial biofilms is mediated by interactions between the cell surface and the substrate

Supplementary information

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## Table S1: Primers used in this study

| fliC_F       | GGAAACCCAATACGTAATCAACGACTTGCAATATAGGATAACGAATC T GTA GGC TGG AGC TG            |
|--------------|---|
| fliC_R       | GAA TAC GGG ACT GCG CTT CAG ACA GGT TGG TAG TGG TGT TG ATA TCC TCC TTA GTT C    |
| fliC_test_F  | CAGACGATAACAGGGTTGAC  |
| fliC_test_R  | TTA CCG GCC TGC TGG ATG AT  |
| csgA_F       | CGT ATT CTC CGG TAG CGC TCT GGC AGG TGT TGT TCC TCA GTA ATG TGT AGG CTG GAG CTG |
| csgB_R       | ${\tt GTACTGATGAGCGGTCGCGTTGTTACCAAAGCCAACCTGAGTCACGTTGACATATGAATATCCTCCTTAG}$  |
| csgAB_test_F | GATGAAACAGTATGGTCAG   |
| csgAB_test_R | GAAGTGACTGTCCATCAG  |
|              |   |



**Figure S1.** Thresholded microscopy image of a competition experiment initiated by linear inoculation of strain MG1655*fimflu*. The sector boundaries from a linear inoculation shows the same chirality as from a droplet inoculation.



**Figure S2.** Competition experiments using *S. pombe* fission yeast and *B. subtilis* bacteria. **a:** Competition experiment between neutral fission yeast strains expressing two different colors. The sector borders are not chiral. **b:** Competition experiment between neutral *B. subtilis* strains expressing different colors. The sector borders are slightly chiral in the same direction as observed for the *E. coli* strains (values of  $\theta$  are given in Table 2).



**Figure S3.** The effect of trypsin. The left panel shows an example of an intensity thresholded image (displayed from the agar side) of colony growth of the *E. coli* MG1655*fimflu* mutant grown on a LB plate with 0.025% trypsin; the scale bar is 1 mm. The right panel show the average boundary angle,  $\langle \phi(r) \rangle$ , (green dotted line), from N=1952 experimental measurements. Also shown is a linear fit (full green line) yielding  $\theta$ =-1.0° (S.E.M=0.005°), the standard error of the mean is shown by a blue punctuated line. Trypsinisation of the MG1655*fimflu* strain significantly reduces chirality to the parental level.