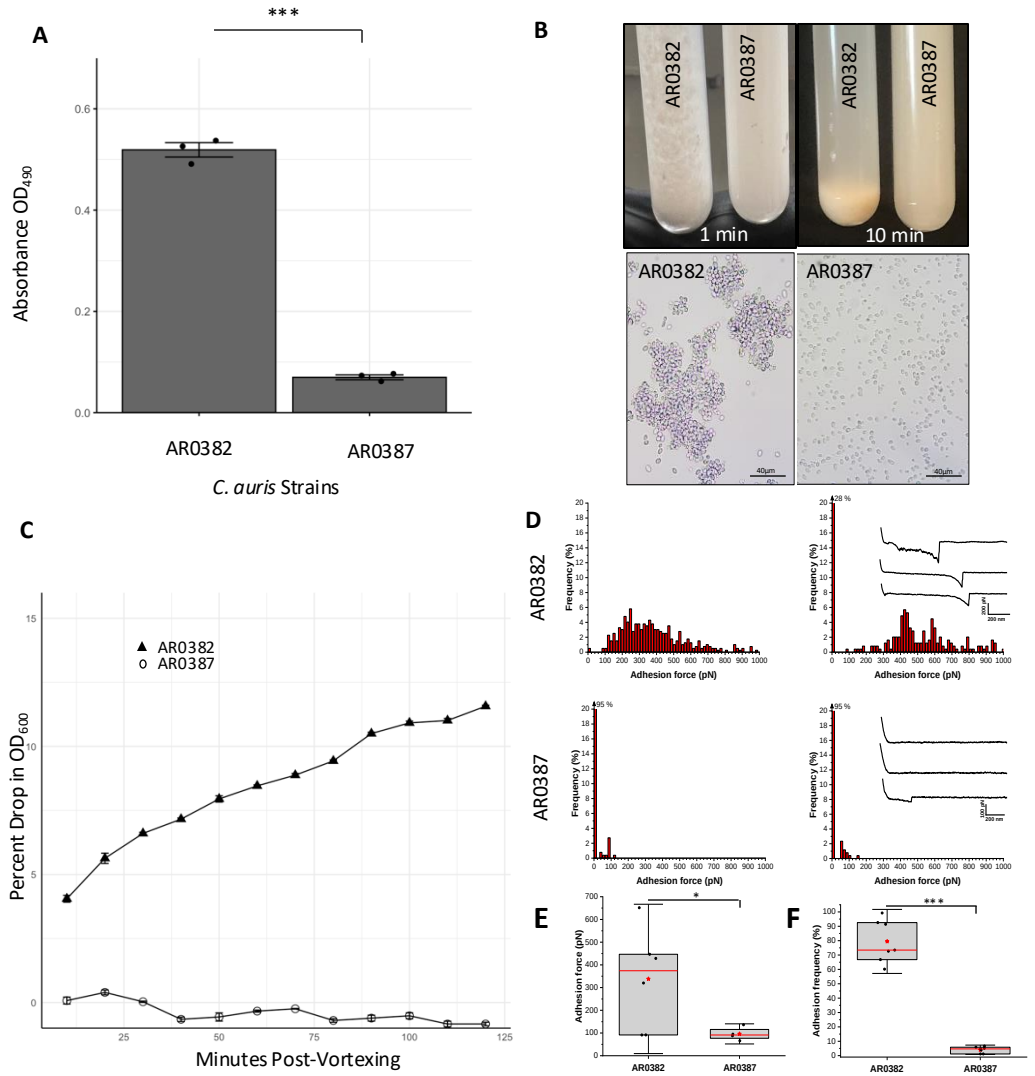
Fig. S1. Comparative evaluation of biofilm formation, aggregation and cell-cell adhesion force by the wild-type AR0382 (aggregative) and AR0387 (non-aggregative) phenotypes. (A) Metabolic activity of 24 h biofilms based on measurements of OD_{490}, optical density. Bar-graphs shows mean and standard error of mean of *n* = 3 biological replicates, each as an average of 4 technical replicates. Statistical analysis was performed by an unpaired two-tailed Welch's t-test. $P = 2.243\times10^{-5}$. **(B)** Aggregation assays, following vigorous vortexing of cell suspensions comparing cell aggregates of AR0382 and AR0387. Bright-field microscopy (lower panel) of aliquots of cell suspensions demonstrating presence of aggregates of AR0382 cells compared to singly suspended cells of AR0387. **(C**) Measurement of rate of cell sedimentation by absorbance readings of OD_{600} of wild-type strains AR0382 and AR0387 over 2 h following vigorous vortexing. Values represent mean OD and standard error of mean of three technical replicates. **(D)** Single-cell force spectroscopy of *C. auris* cell-cell adhesion. Adhesion force histograms with representative retraction profiles (inset) obtained for the interaction between AR0382 wild-type cells and the interaction between AR0387 cells; 2 representative cell pairs are shown for each strain. **(E)** Adhesion force boxplots depicting *n* = 6 and *n* = 4 cell pairs for AR0382 and AR0387 respectively. Statistical analysis was performed by an unpaired two-tailed Welch's t-test. $P = 4.21\times10^{-2}$ **(F)** As in **(E)**, adhesion frequency boxplots show interactions between *n* = 7 cell pairs for both strains. $P = 8.06\times10^{-6}$. Red stars represent the mean values, red lines are the medians, boxes are the 25−75% quartiles and whiskers the standard deviation from mean. *0.01 < *P* ≤ 0.05, ***$P < 0.001$.



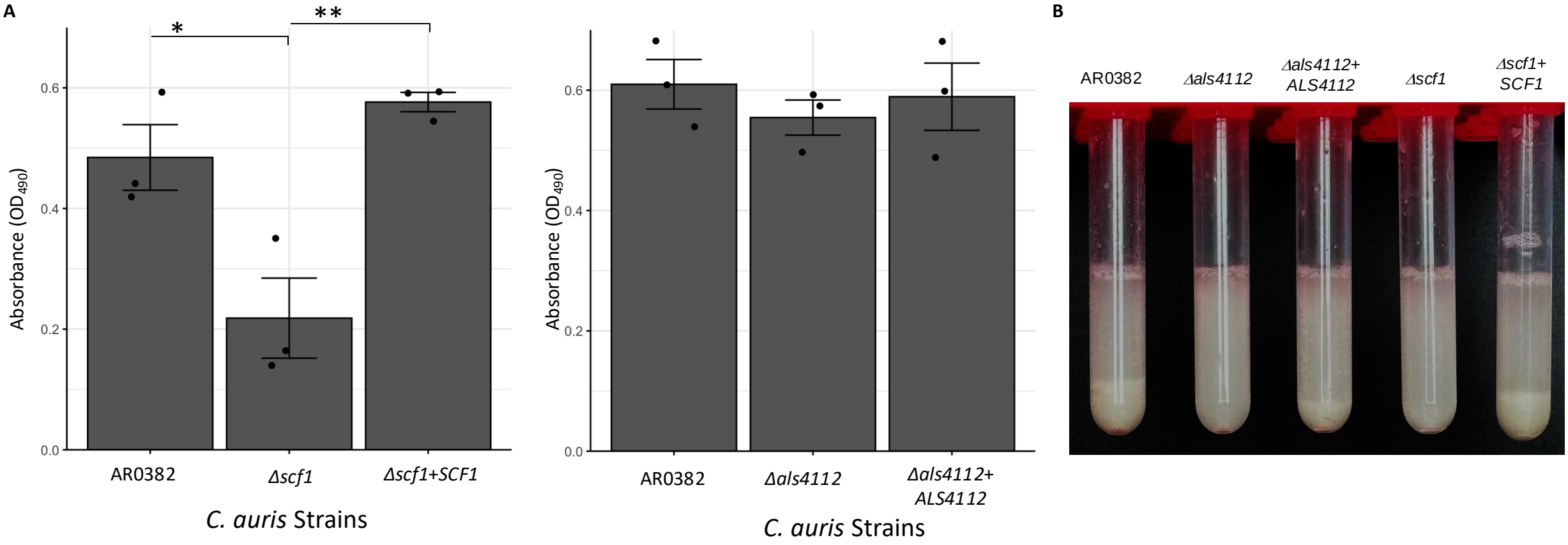


Fig. S2. Comparative evaluation of biofilm formation and aggregation by complemented strains of mutants of *ALS4112* and *SCF1* genes (Δ*als4112+ALS4112 and* Δ*scf1+SCF1)* to their respective mutants and wild-type strain. (A) Metabolic activity of 24 h biofilms based on measurements of OD_{490}. Bar-graphs shows mean and standard error of mean of $n = 3$ biological replicates, each as an average of 4 technical replicates. Statistical analysis was performed by one-way ANOVA and post-hoc Tukey test with p-values representing significant differences. $P = 2.25\times10^{-2}$, 5.72×10^{-3} ** $0.001 < P \leq 0.01$, ***$P < 0.001$. **(B)** Cell aggregation following 2 min of vigorous vortexing.

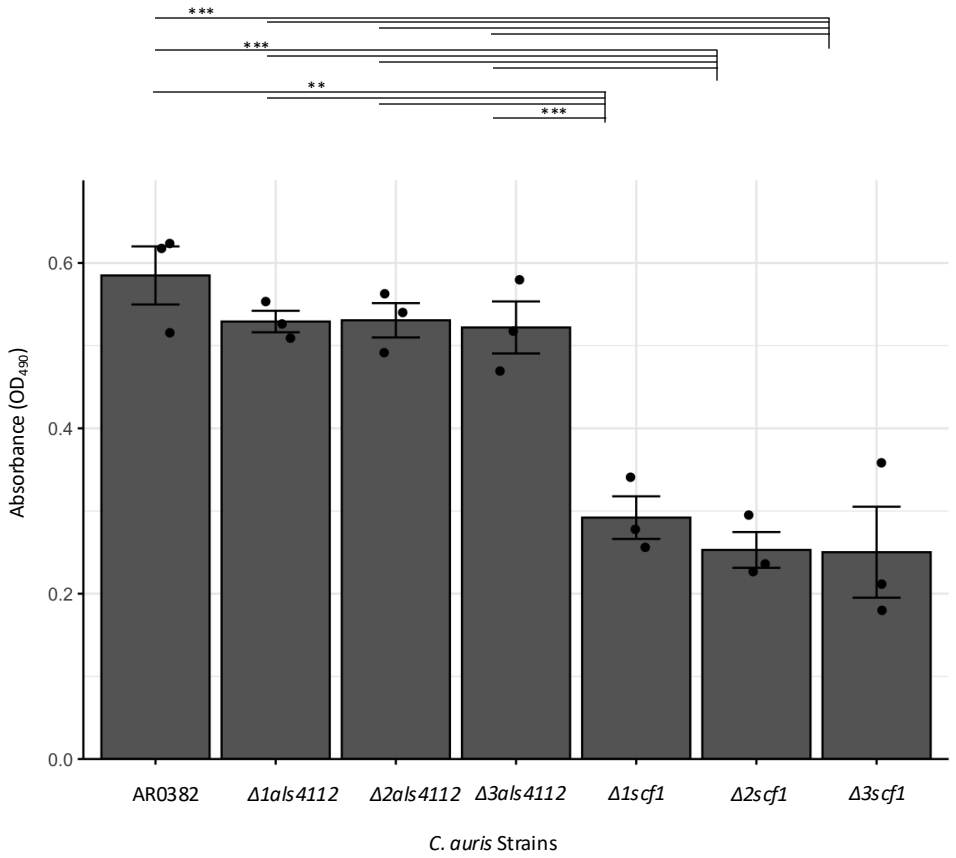


Fig. S3. Evaluation of biofilm formation by the 3 mutant strains generated for the *ALS4112* and *SCF1* genes (*Δ*1-*Δ*3). A measurement of the metabolic activity of 24 h biofilms based on values of OD_{490} comparing all generated mutant strains to the wild-type. Boxplots show mean and standard error of mean of $n = 3$ biological replicates, each as an average of 4 technical replicates. Statistical analysis was performed by one-way ANOVA and post-hoc Tukey test with p-values representing significant differences. $P = 1.64\times10^{-3}$, 1.55×10^{-3}, 2.18×10^{-3}, 2.02×10^{-4}, 3.70×10^{-4}, 3.50×10^{-4}, 4.83×10^{-4}, 5.17×10^{-5}, 3.35×10^{-4}, 3.17×10^{-4}, 4.37×10^{-4}, 4.71×10^{-5} **$0.001 < P \leq 0.01$, ***$P < 0.001$.

Supplementary Table 1 (S1). Primers used in this study.

Marker	Purpose	Primer name	Sequence (5'-3')
B9J08_004112	US_cloning	CauALS5_US_GIB_F	AACTTCCTCGAGGGGGGGCCGAAAGATGATGGGAAACAAGGTGAAG
	US_cloning	CauALS5_US_GIB_R	AGGGAACAAAAGCTGGGTACCTCACCACGAGACGGGAGC
	DS_cloning	CauALS5_DS_GIB_F	GCGAATTGGAGCTCCACCGCGGTCCCCAGGTGCTATTTCTTG
	DS_cloning	CauALS5_DS_GIB_R	AGATCCACTAGTTCTAGAGCACGTGAGCTTTTATGATACCTAC
	crRNA	ALS5_crRNA	GTACTCAGGCGAAAACATCG
		ALS5US_chF	GTCATTCTTCCTTGTTTCCG
		ALS5DS_chR	CATTTGCAGAGAAGAAATCTGCGC
B9J08_001458	US_cloning	Gbson_RBT1US_F	TAGAAAGTATAGGAACTTCCTGTGGAGGTGAAGTTTTAAGATAGAG
	US_cloning	Gbson_RBT1US_R	AGGGAACAAAAGCTGGGTACGCTCGCCGCTCACAATG
	DS_cloning	Gbson_RBT1DS_F	CTATAGGGCGAATTGGAGCTGTCGGGATTGTGGGAATTC
	DS_cloning	Gbson_RBT1DS_R	AGATCCACTAGTTCTAGAGCTTCTAATGACTGATACTCATACTTTC
	Verify deletion	RBT1US_chF	ATGTGCTTCTTCTGGGTCTTTTG
	Verify deletion	RBT1DS_chR	GCGATAGGAGACGATGTTGATAAC
	crRNA	RBT1_crRNA	CTAGGTCCACTAGGTCCACT
pSFS2	PCR/Seq/verify deletion	pSAT1_US-cPCR_F	CTAACGATGCATACGACTACATC
	PCR/Seq/verify deletion	pSAT1_DS-cPCR_R	ACATATGTGAAGTGTGAAGGGGG