Table S1. Confluence [%] of analyzed staphylococcal biofilms (number of strains = 10)

strain description	confluence [%]	average confluence [%] for n=10 strains
ATCC 6538	99,9	93,61±5,55
ATCC 33591	99,9	
S1	81,7	
S2	95,7	
S3	94,4	
S4	97,8	
S5	95,3	
S6	91,2	
S7	90,8	
S8	89,4	

Table S2. The Fluorescence Intensity of live and dead (L, D, respectively) staphylococcal biofilms with regard to their location in **T** (top), **M** (middle) and **B** (bottom) parts of biofilm

Strain no	**Part of biofilm**	**Fluorescence Intensity L:D**	**Matches pattern FI M > FI T and FI B?**
ATCC 6538	T	980: 710	**Yes**
	M	2499:1441	
	B	258: 742	
ATCC 33591	T	492: 938	**Yes**
	M	891: 689	
	B	161:591	
S1	T	664:523	**Yes**
	M	1725:1705	
	B	326:623	
S2	T	1353:779	**Yes**
	M	4130:3174	
	B	703:1021	
S3	T	954:627	**Yes**
	M	4265:3094	
	B	498:295	
S4	T	769:412	**Yes**
	M	1900:621	
	B	219:492	
S5	T	205:474	**Yes**
	M	432:381	
	B	176:321	
S6	T	522:634	**Yes**
	M	1421:839	
	B	283:425	

S7	T	405:174	**Yes**
	M	571:331	
	B	108:273	
S8	T	411:290	**Yes**
	M	710:629	
	B	138:305	

Figure S1. Three types (A,B,C) of distribution of dead/cell wall compromised cells vs. viable/cell wall non-compromised cells in staphylococcal biofilm *in vitro*; the vertical cross-section through the biofilm structure. A – strain ATCC 6538; B – strain S1, C -ATCC 33591. Scale bar is 30 μm. Microscope SP8, magn.40x.

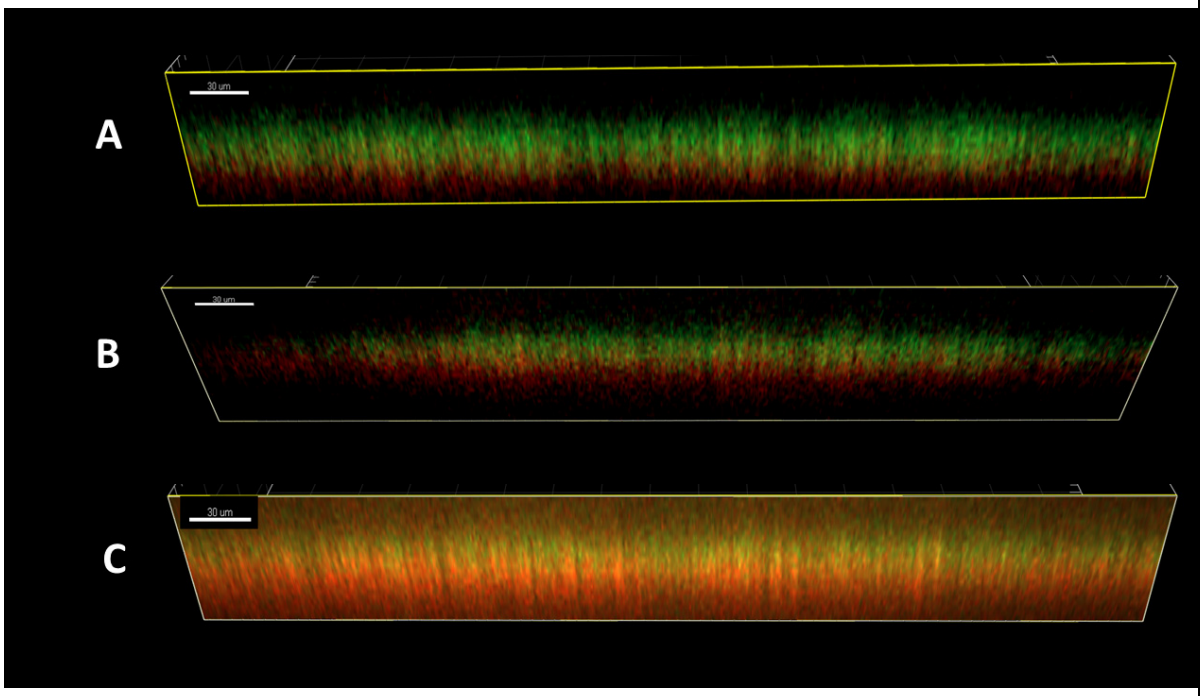
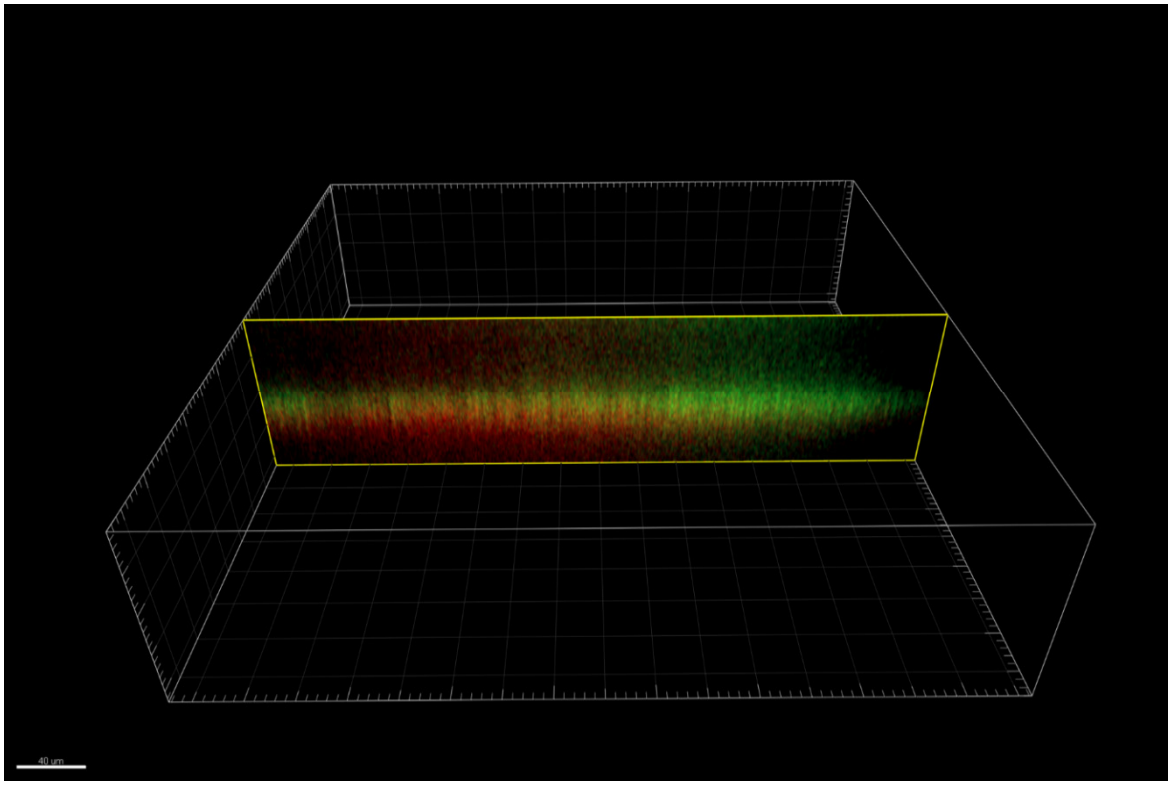


Figure S2. The different types of Live/Dead cells' distribution in biofilm within single plate. Strain ATCC 33591. Scale bar is 40μm



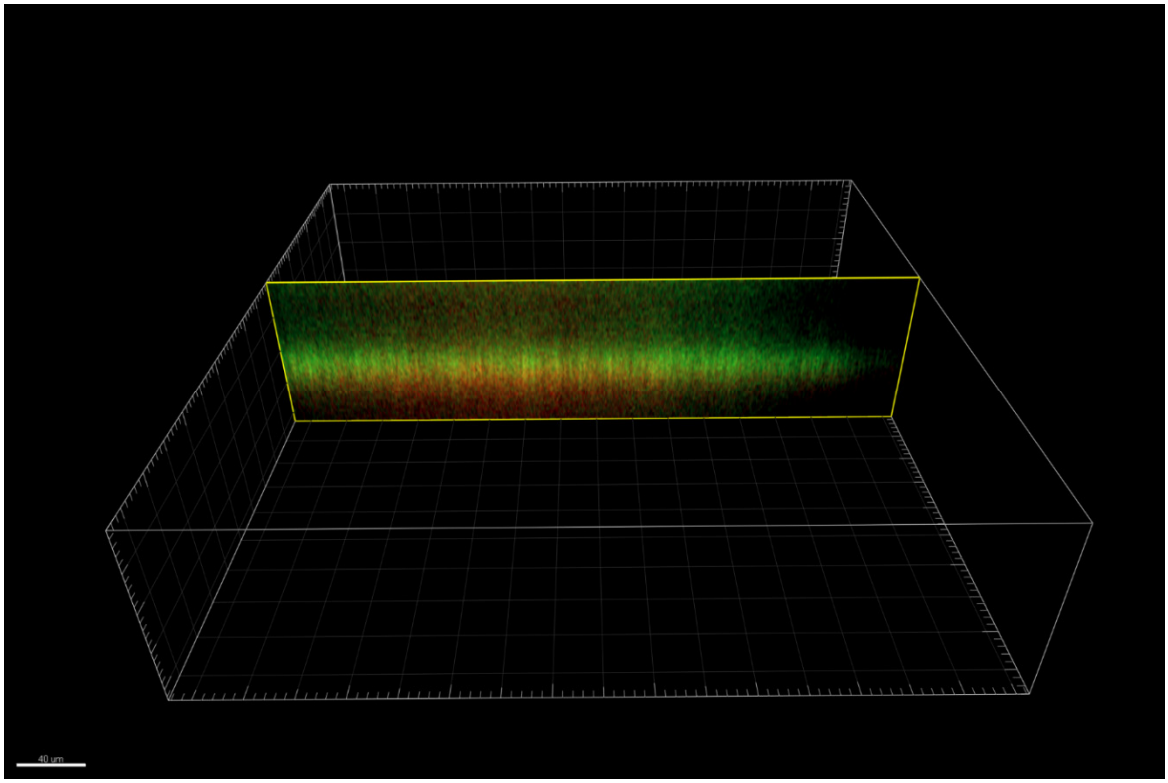


Figure S3. The impact of operators' various pipetting habits (A,B,C) on dye distribution in the well (of a 24-well plate) covered with biofilm formed by the same staphylococcal strain (ATCC 6538). Picture D presents the biofilm of the aforementioned strain dyed with the L/D method by an operator with 3 years of experience in staphylococcal biofilm culturing and dyeing. The red cross indicates the approximate place of pipette tip placement.