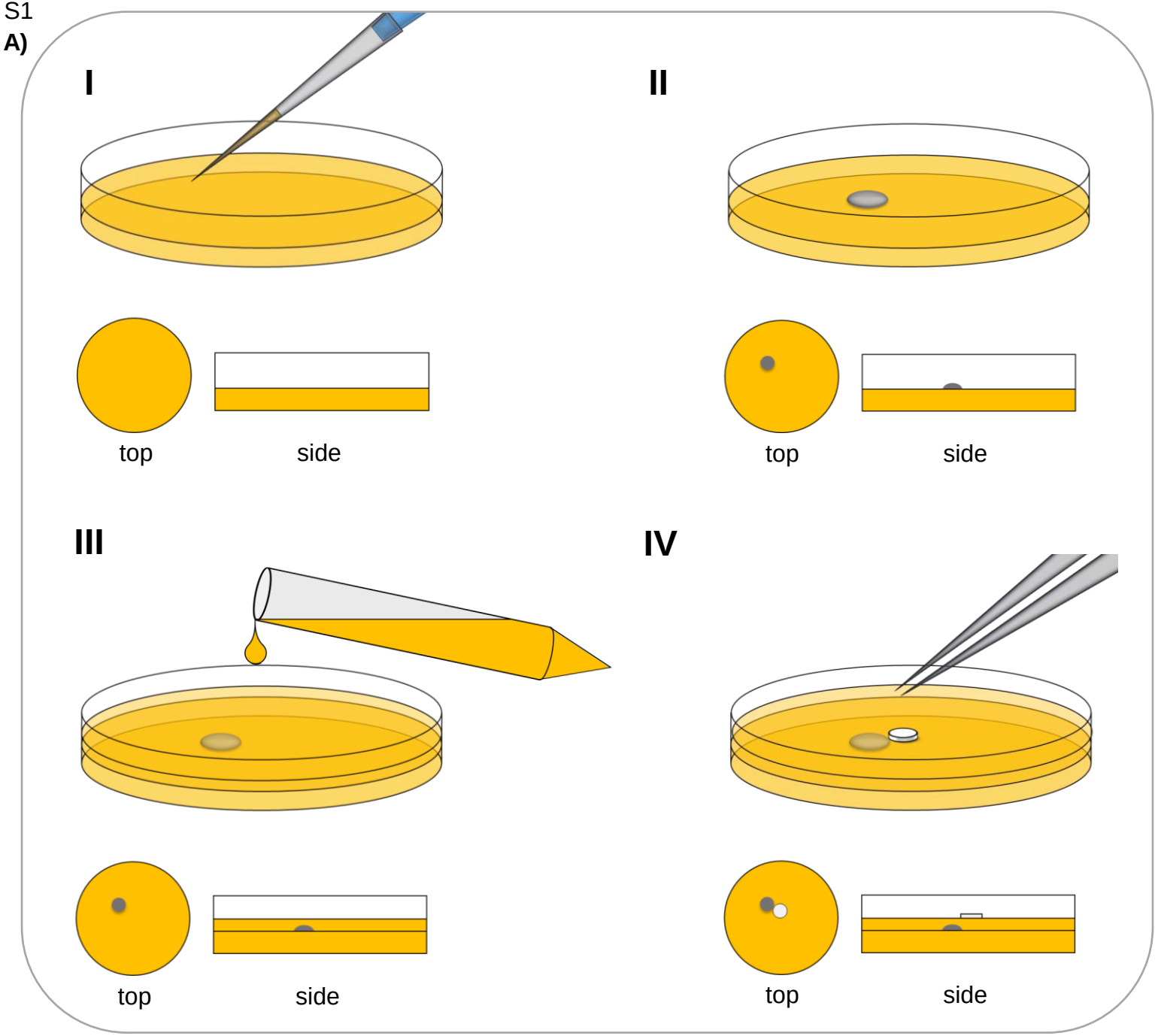
Fig. S1

A)


I
top
side
II
top
side
III
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side
IV
top
side

B)

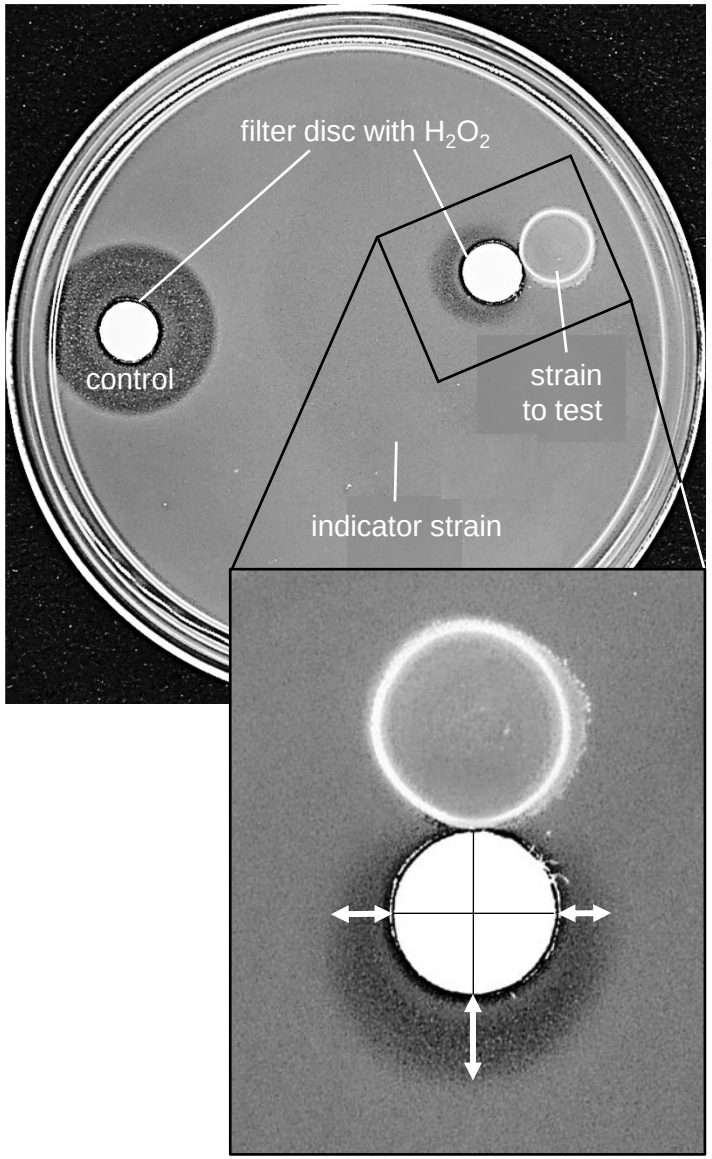
filter disc with H_2O_2
control
strain
to test
indicator strain

Fig. S1: H_2O_2 protection assay. (A) Schematic display: (I) Application of strain to be tested for its protective capacity. (II) Situation after 24 hours of incubation. (III) Overlay with soft agar containing an indicator strain. (IV) Application of a filter disc containing H_2O_2 onto solidified soft agar in close proximity to the "colony" of the tested strain and subsequent incubation for 24 hours. For better understanding, agar plates of all working steps are shown as additional top and side views. (B) Exemplarily, shown is an agar plate like it appears after 24 hours of incubation with H_2O_2 discs. The magnified section of the image shows the lateral and distal sites of measurement of the inhibition zones (white arrows).

Supplemental Material - Pyruvate Secretion by Oral Streptococci Modulates Hydrogen Peroxide Dependent Antagonism
Sylvio Redanz [1]*, Puthayalai Treerat [1], Rong Mu [1], Ulrike Redanz [1], Zhengzhong Zou [1], Dipankar Koley [2], Justin Merritt [1,3], and Jens Kreth [1,3]
[1] Department of Restorative Dentistry, Oregon Health & Science University, Portland, OR 97239, USA; [2] Department of Chemistry, Oregon State University, Corvallis, Oregon 97331, USA; [3] Department of Molecular Microbiology and Immunology, School of Medicine, Oregon Health & Science University, Portland, OR 97239, USA

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Fig. S2

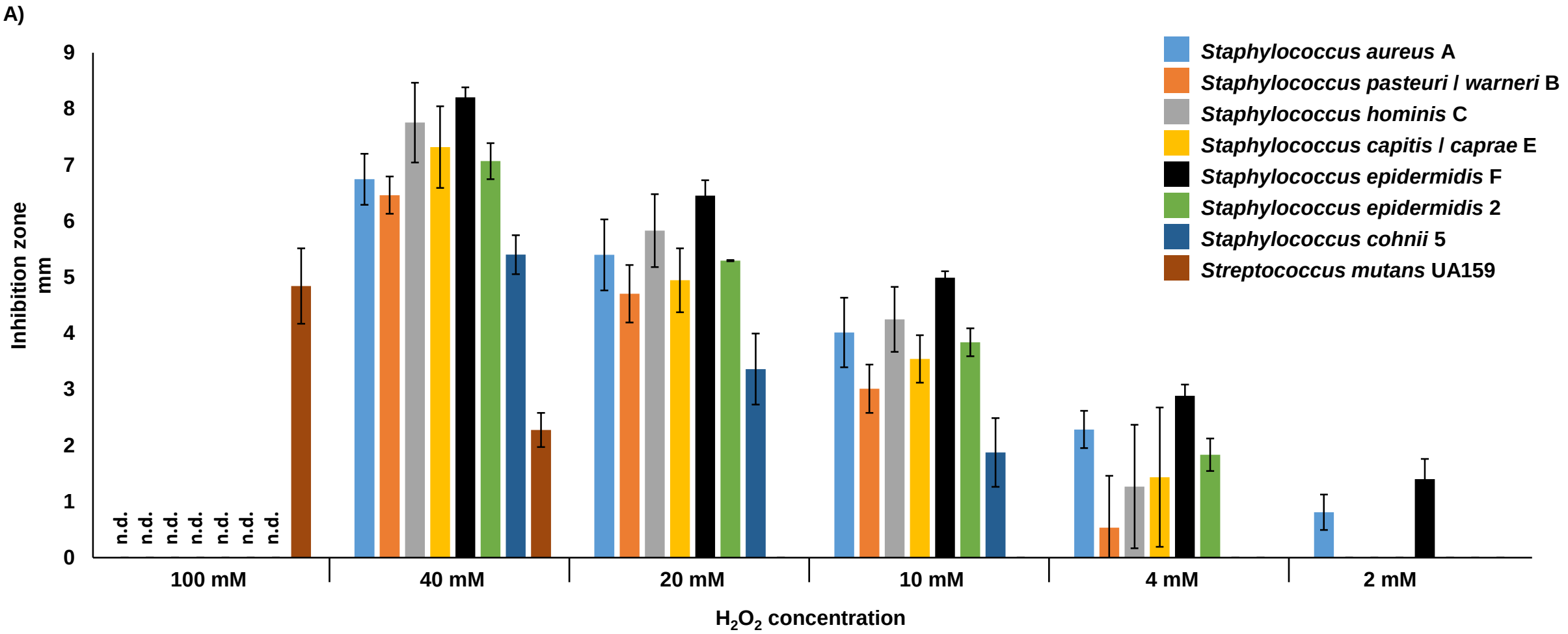
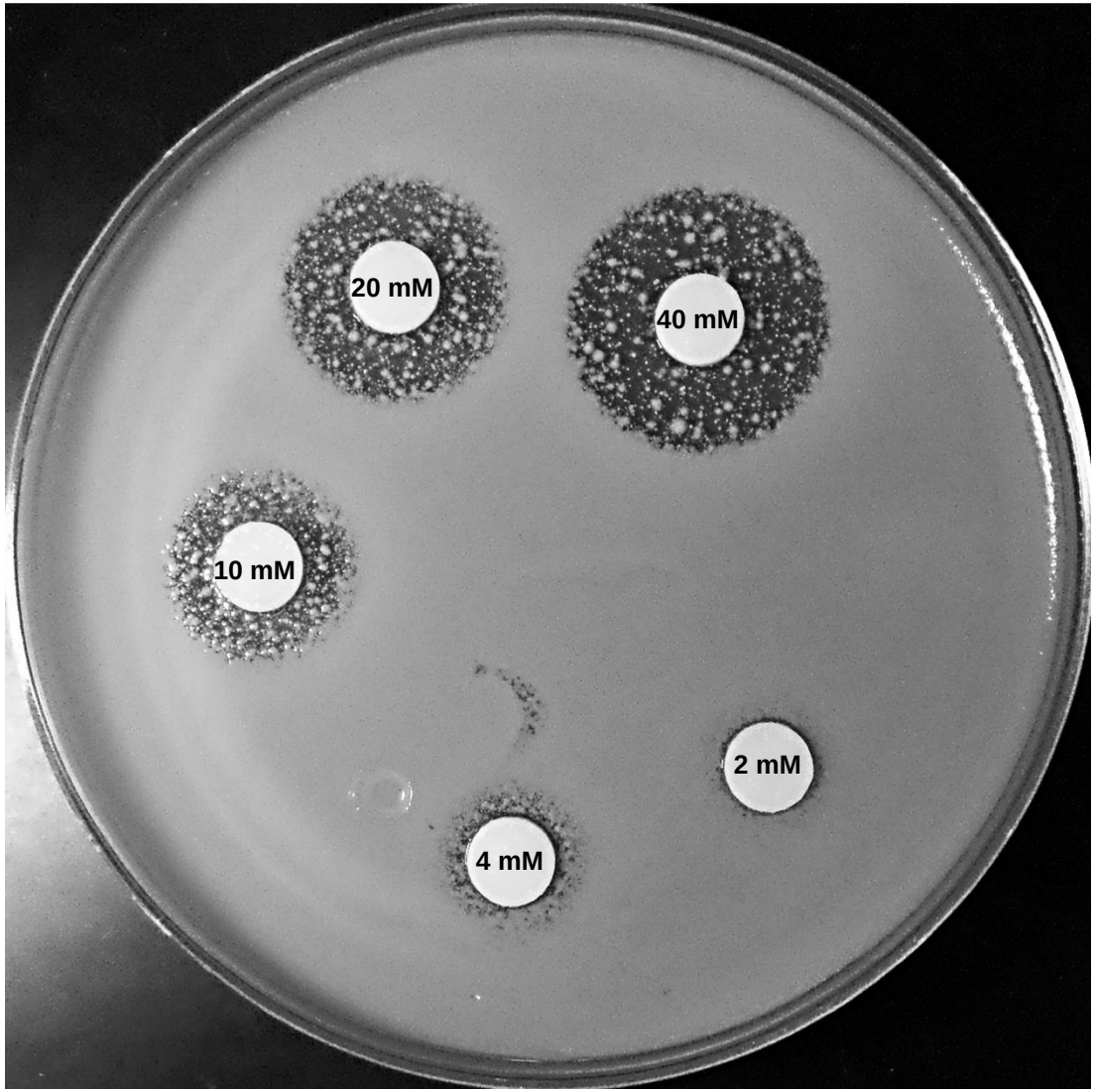


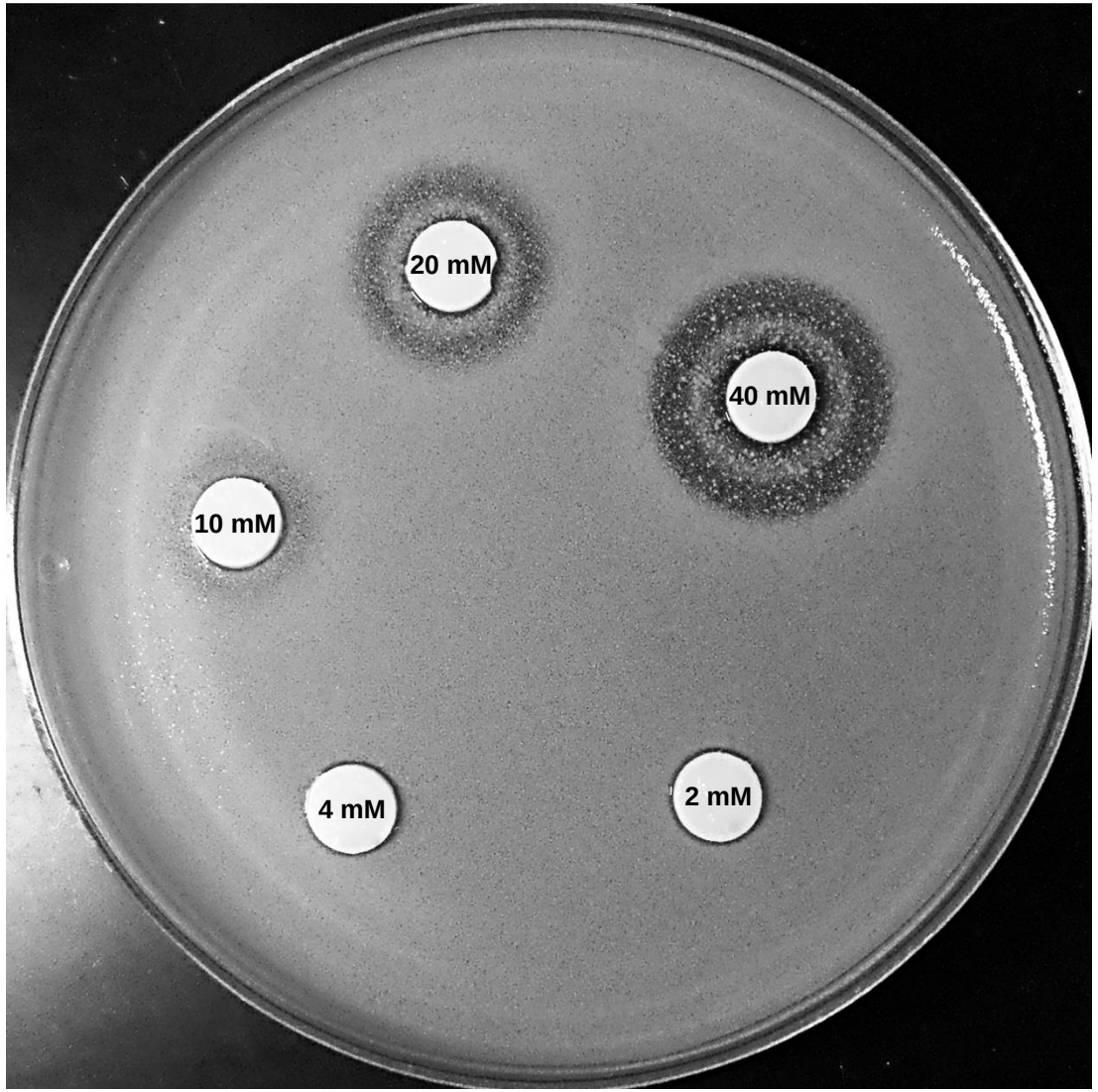
Fig. S2: Comparison of indicator species. H_2O_2 protection assay were performed as described in the main text. Briefly, solidified soft agar (60 °C) was inoculated with the indicated indicator strains and poured on a BHI (bacto) agar plate. After 30 minutes at room temperature (solidification of soft agar) filter discs were soaked with 40 µl fo H_2O_2 solutions with indicated concentrations. (A) Shown are means and standard deviations of three independent experiments. (B) The following slides display representative examples. n.d. – not determined

Fig. S2

B)



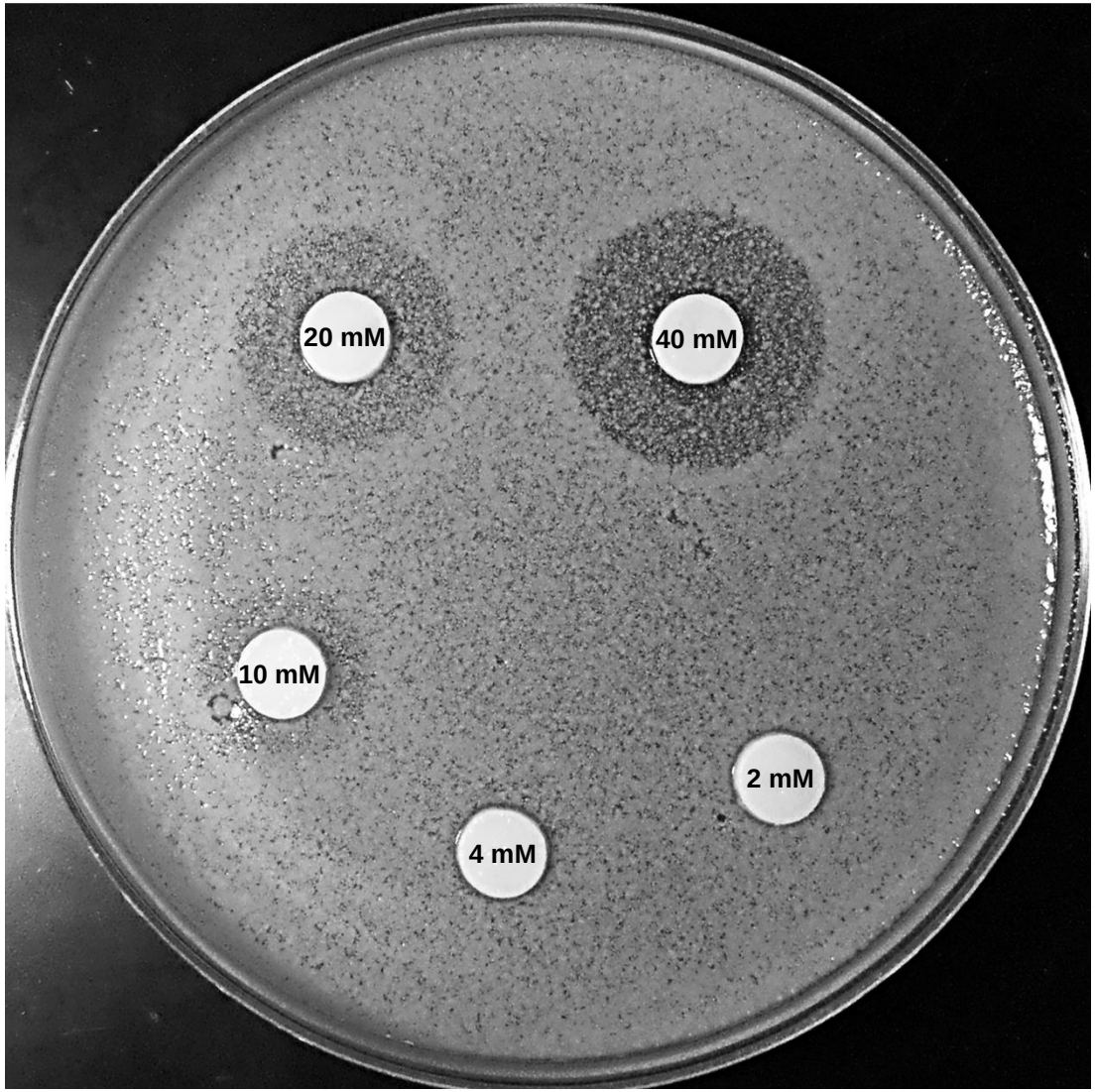
***Staphylococcus aureus* A**



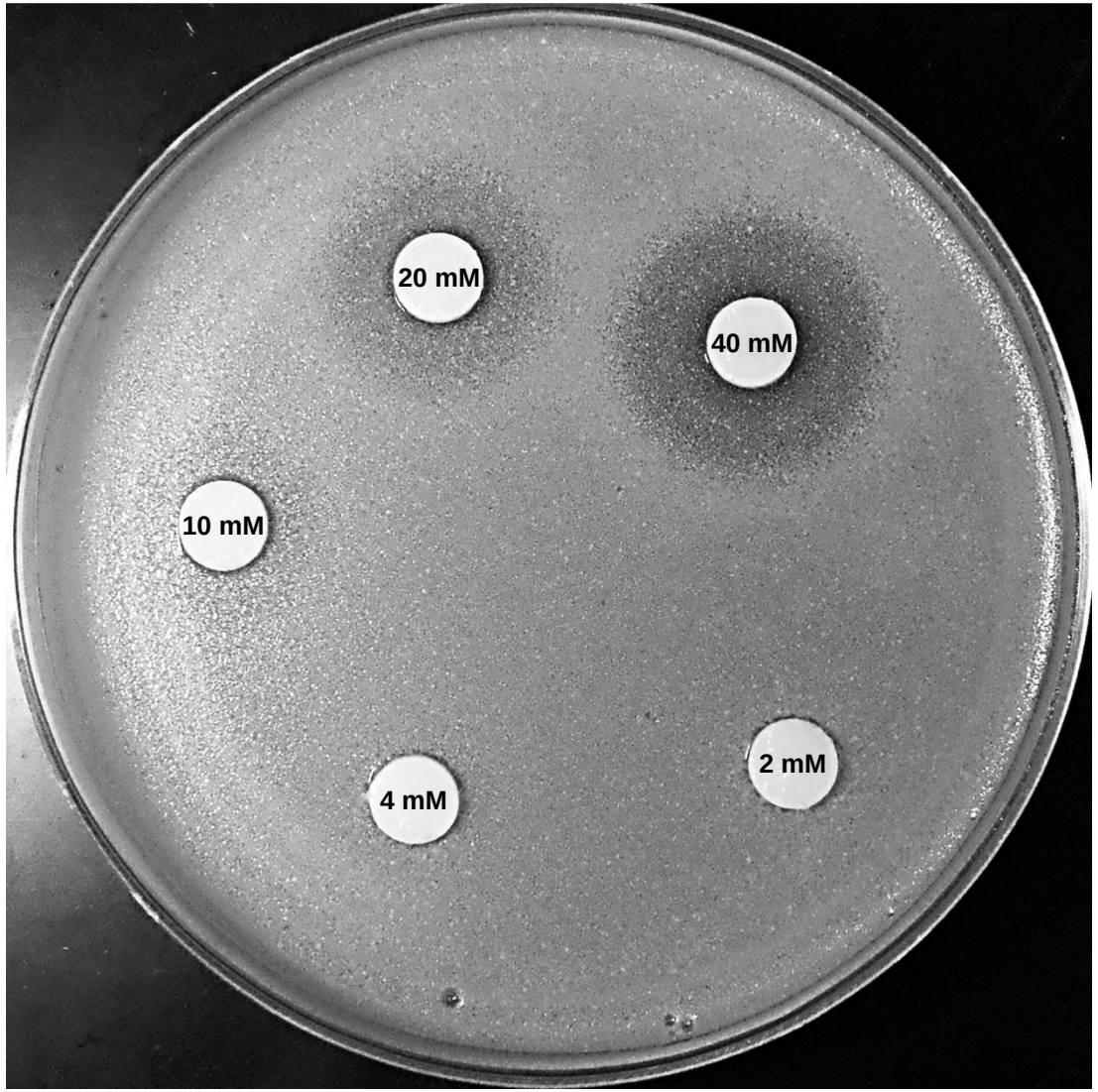
***Staphylococcus pasteuri / warneri* B**

Fig. S2

C)



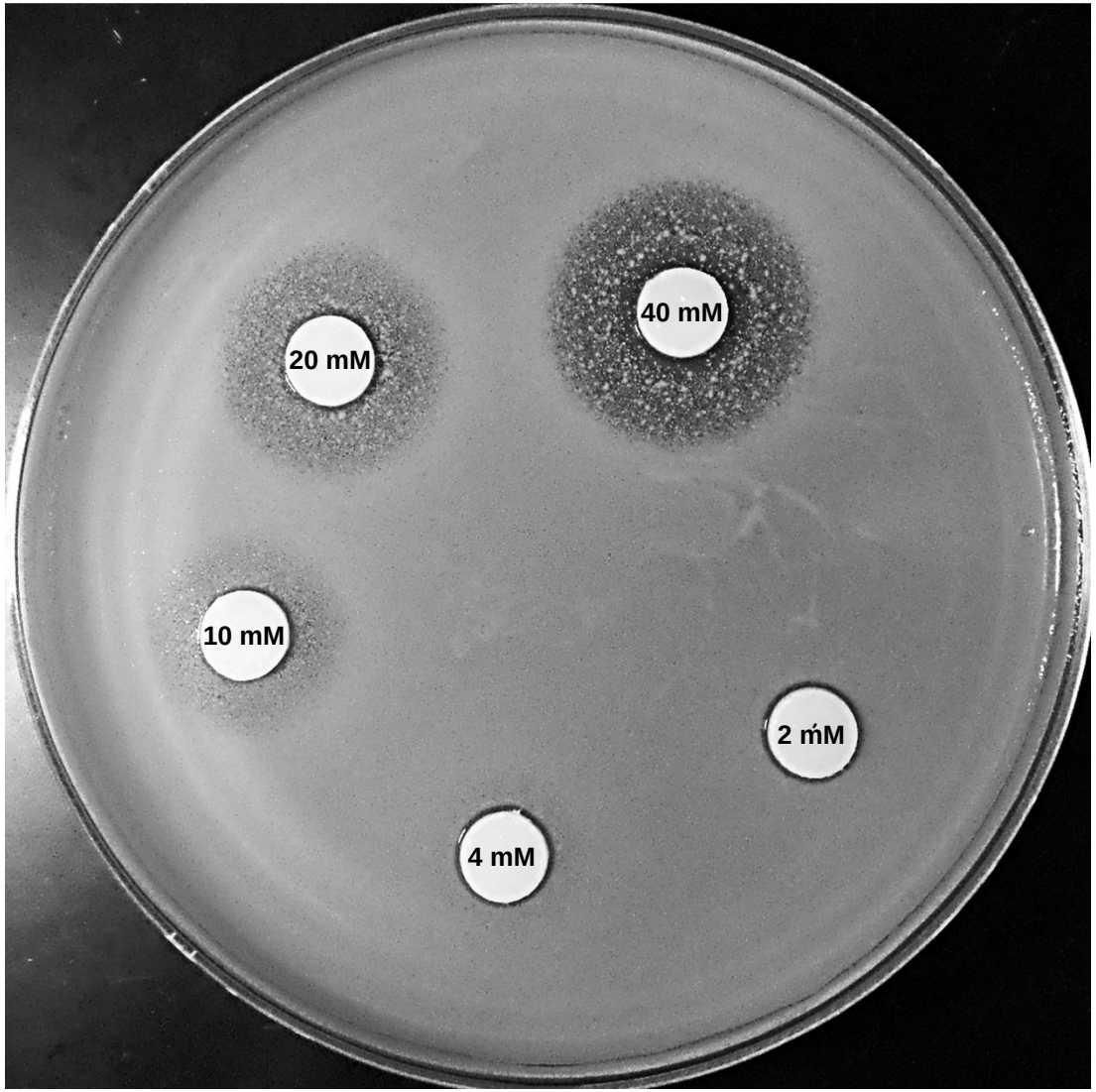
***Staphylococcus hominis* C**



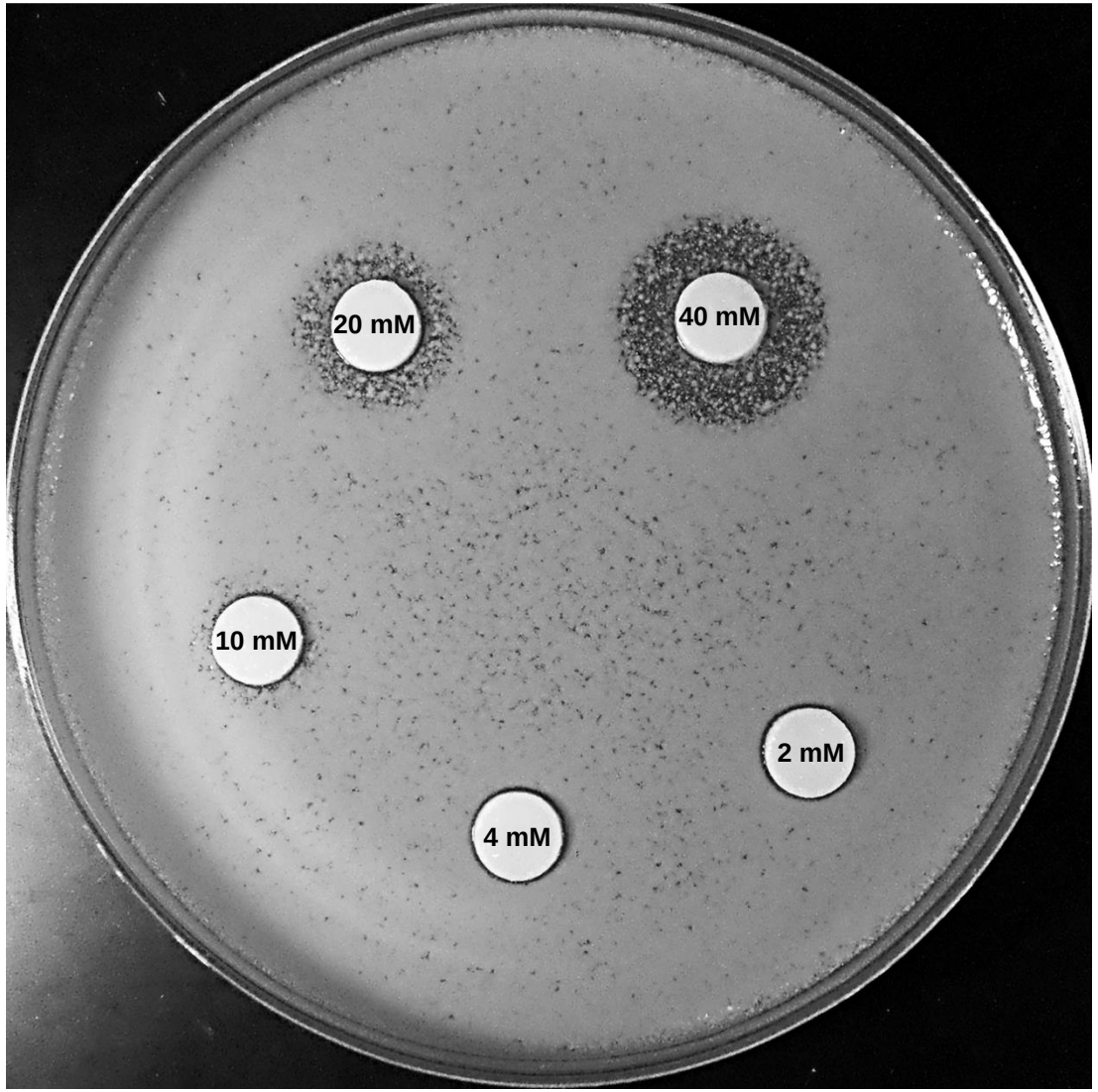
***Staphylococcus capitis / caprae* E**

Fig. S2

D)



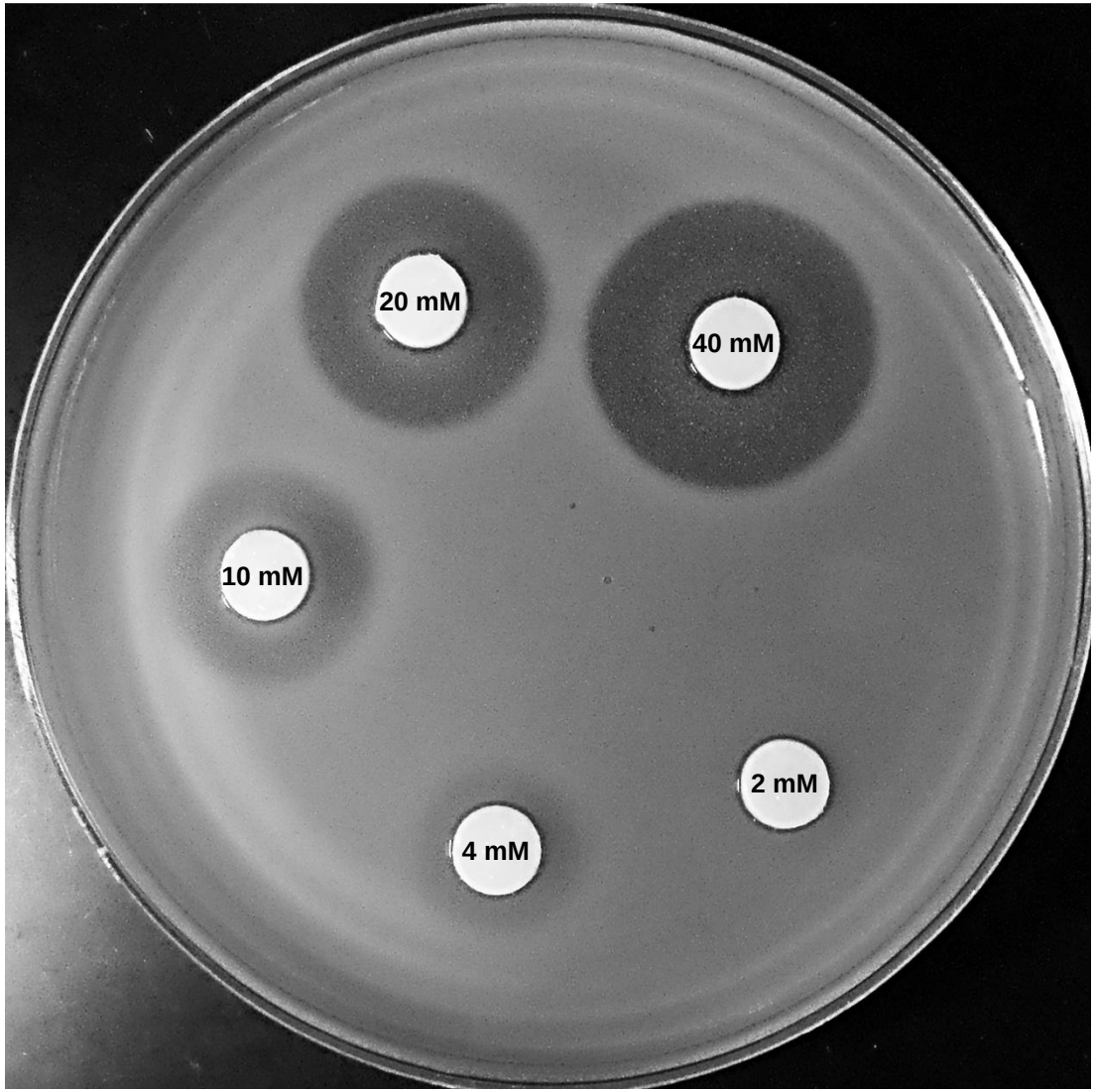
***Staphylococcus epidermidis* 2**



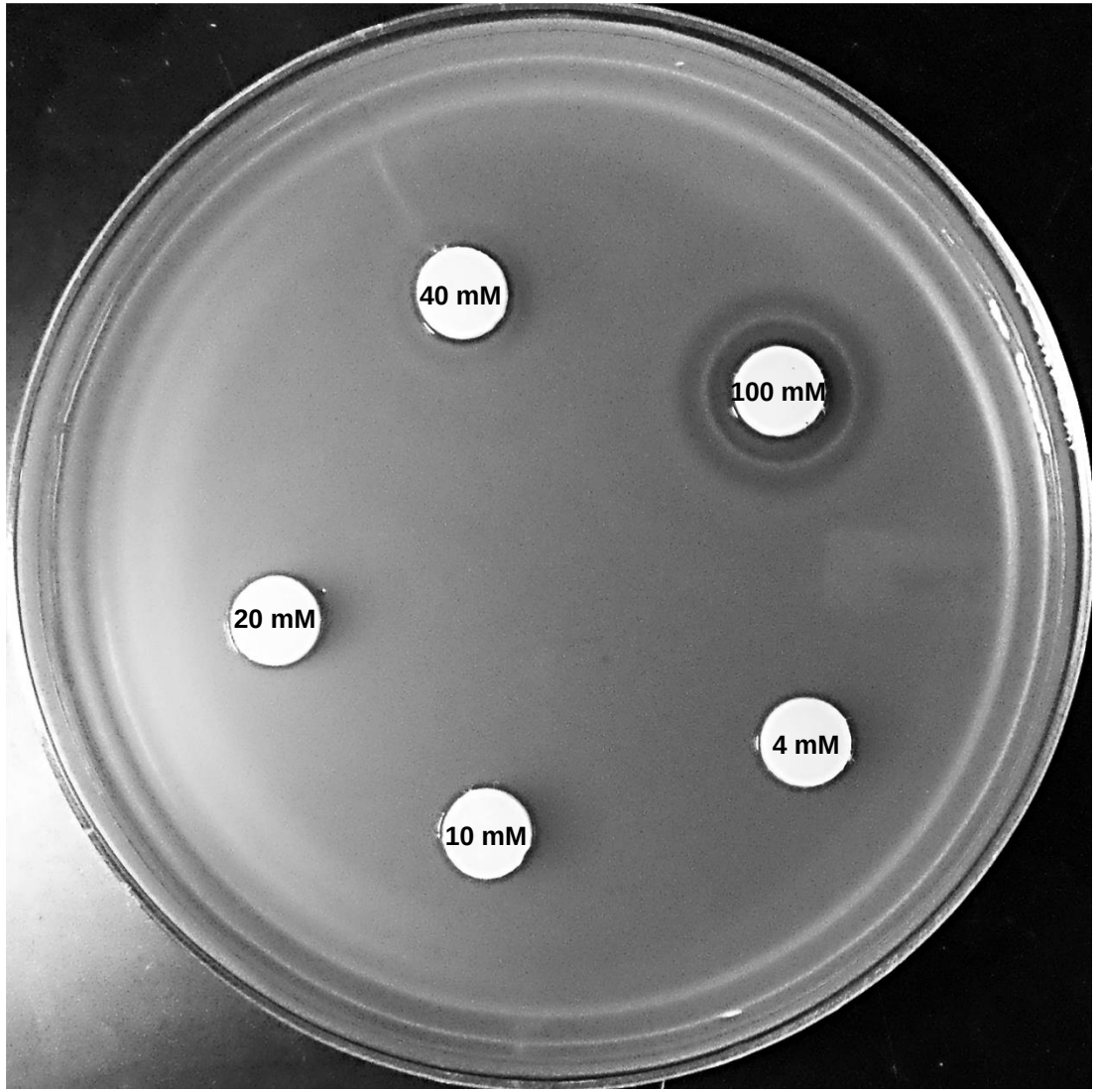
***Staphylococcus cohnii* 5**

Fig. S2

E)



***Staphylococcus epidermidis* F**



***Streptococcus mutans* UA159**

Fig. S3

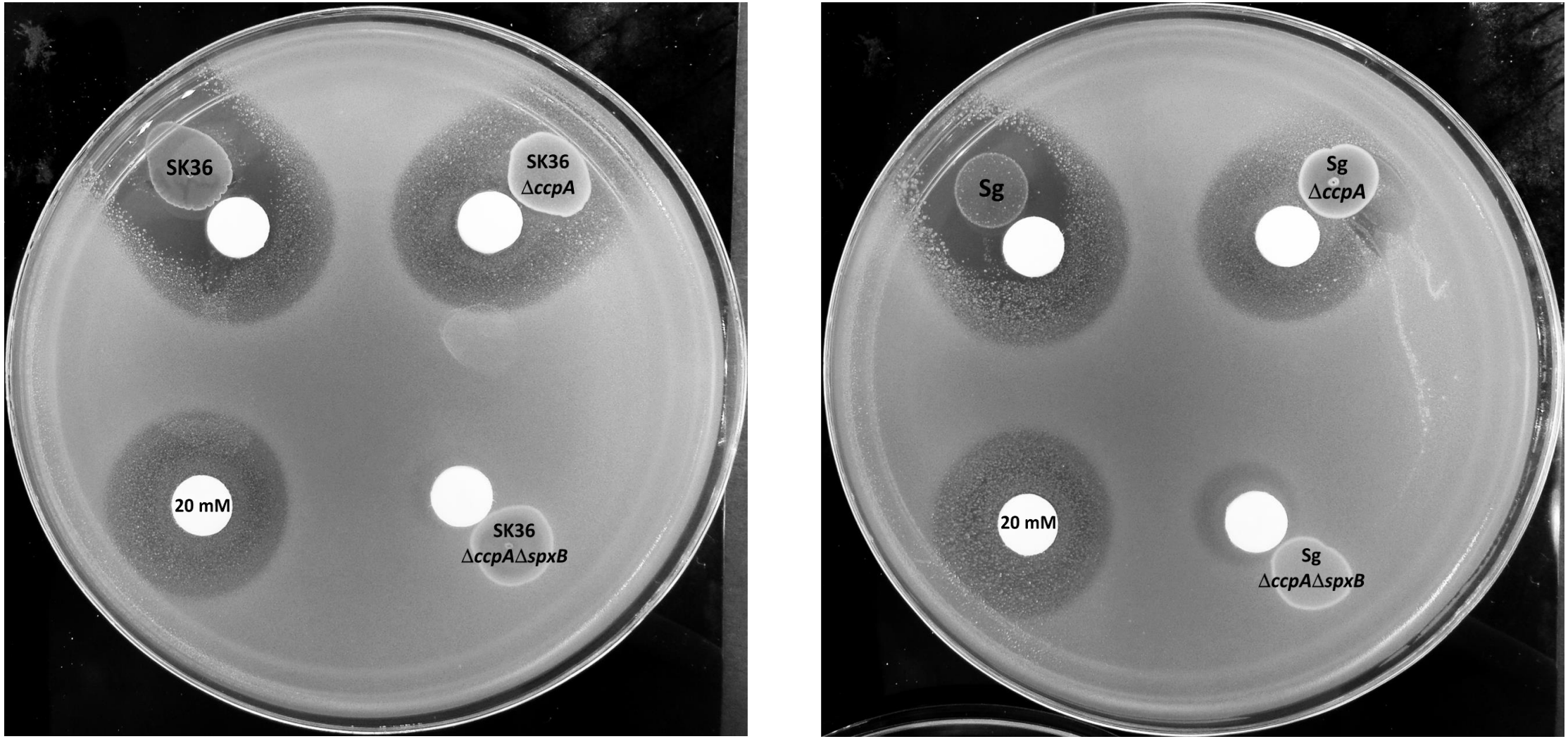


Fig. S3: *S. sanguinis* SK36 and *S. gordonii* DL1 derivatives tested in an H_2O_2 protection assay. SK36 and DL1 derivatives were grown on BHI agar plates, overlaid with soft agar containing *S. epidermidis* F (indicator strain), and challenged with 20 mM H_2O_2 (40 µl) delivered by previously soaked filter discs. The inhibition zone around the filter disc in lose proximity to the *spxB*/*ccpA* double mutants of *S. sanguinis* SK36 (left) and *S. gordonii* DL1 (right) is significantly reduced compared to the control disc, demonstrating the protective effect. Inhibition zones in close proximity to the wildtypes and isogenic ccpA mutants are almost unaffected due to the autologous release of H_2O_2 in these strains (significant inhibition zones appear around the tested strains). Shown are representative plates of the assay.

Fig. S4

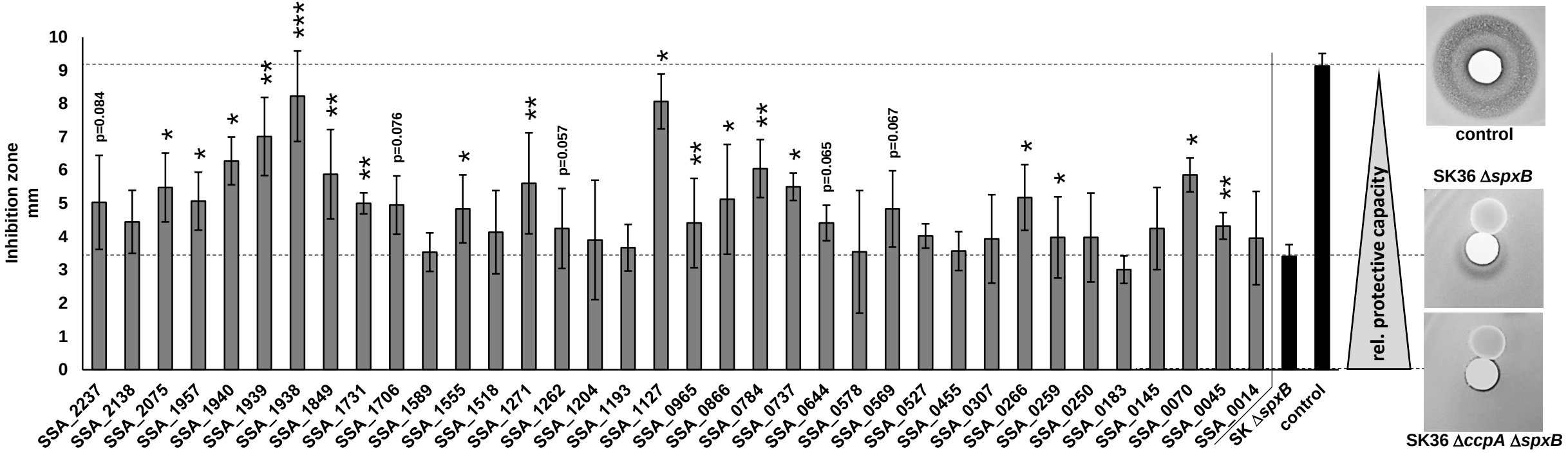


Fig. S4: H_2O_2 protection assay with transposon mutants. H_2O_2 protection assays have been performed with the indicated mutants and controls. Shown are the inhibition zone sizes of all tested transposon mutagenesis based mutants in the SK Δ*spxB* background, which passed a two step screening. Displayed are averages and standard deviations of three independent experiments. Paired t-tests (two-tailed) were performed by testing each mutant against the respective control on the same plate. Please note, the displayed controls are just shown for better visualization and represent averages of all performed experiments. Dashed lines indicate the average size of inhibition zones of SK36 Δ*ccpA*/Δ*spxB*, SK36 Δ*spxB* and the control shown as reference; representative pictures are shown on the right. Significance levels: *p<0.05; **p<0.01; ***p<0.001 (For non significant data sets with a p-value below 0.1, the respective p-values are given.)