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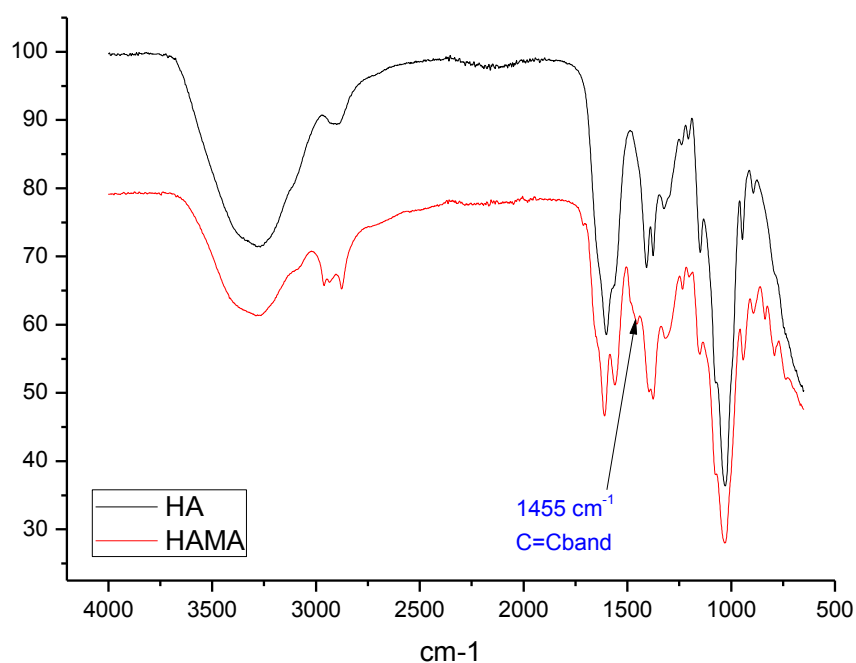
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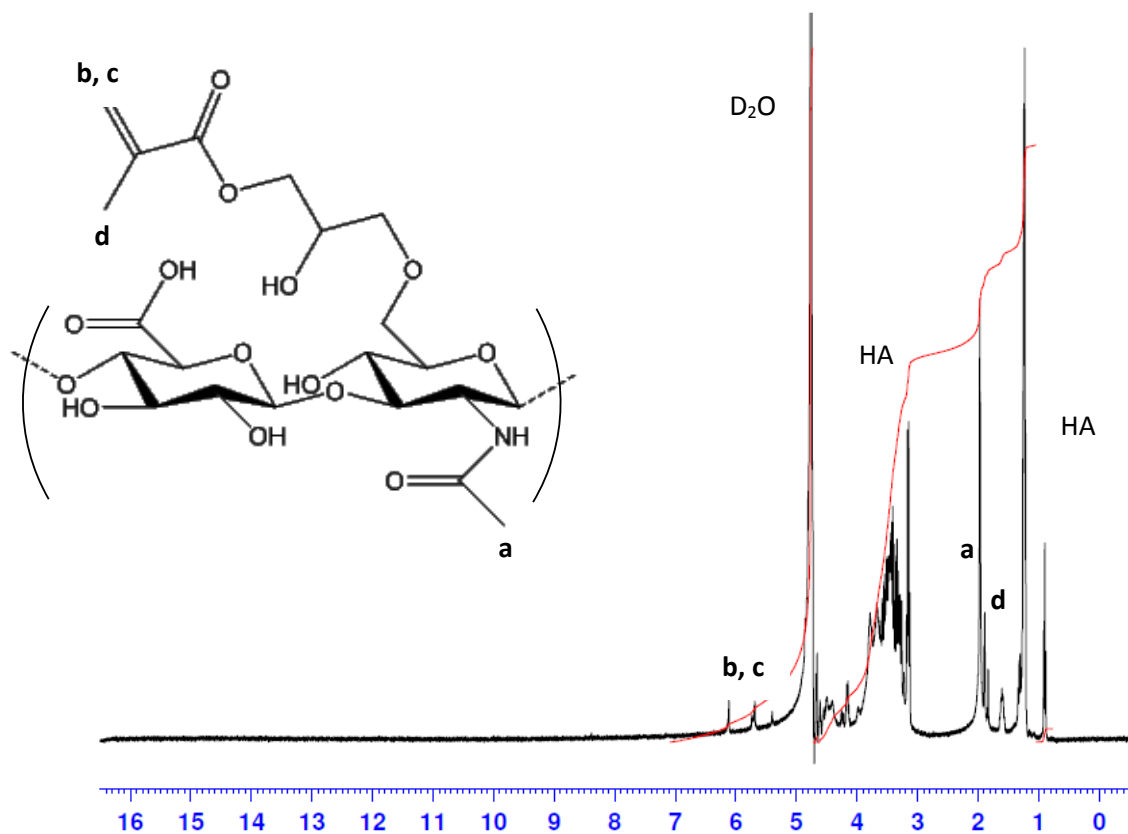
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## SUPPORTING INFORMATION Triggered Release of Bacteriophage K from Agarose/Hyaluronan Hydrogel Matrices by *Staphylococcus aureus* Virulence Factors

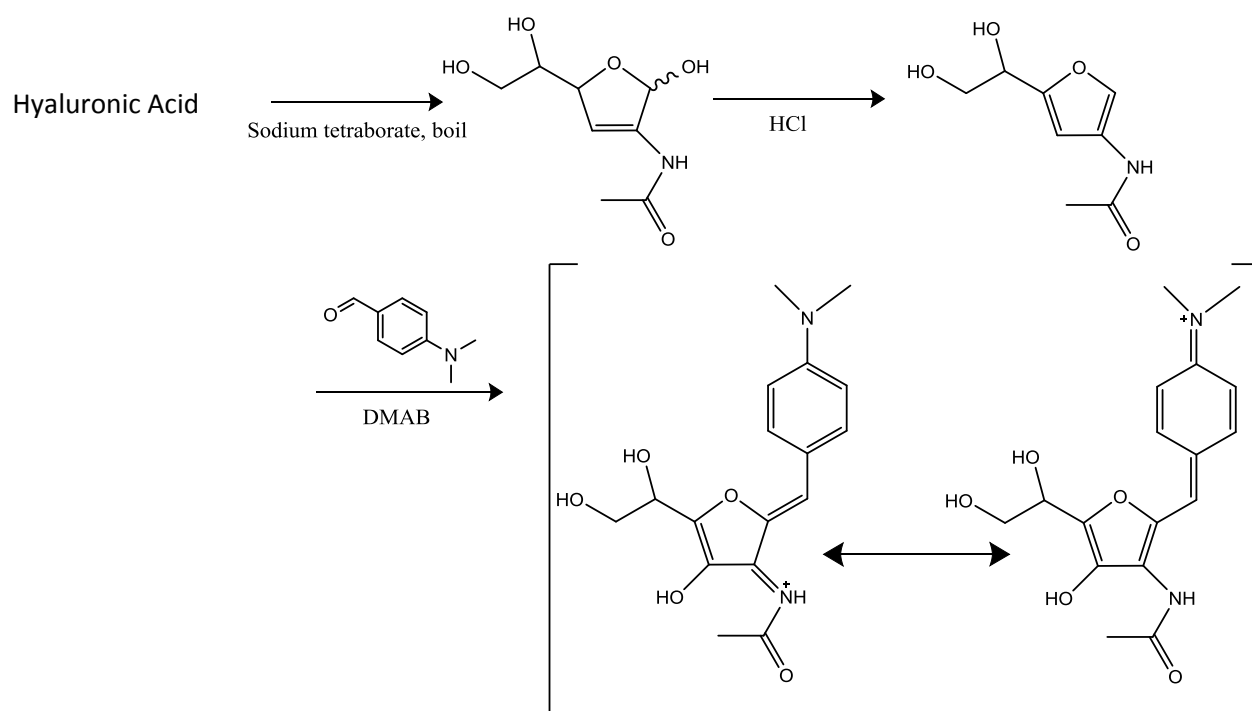
Jessica E. Bean<sup>1</sup>, Diana R. Alves<sup>1</sup>, Maisem Laabei<sup>1</sup>, Patricia P. Esteban<sup>2</sup>, Naing Tun Thet<sup>1</sup>, Mark C. Enright<sup>1</sup>, A. Toby A. Jenkins<sup>1\*</sup>.



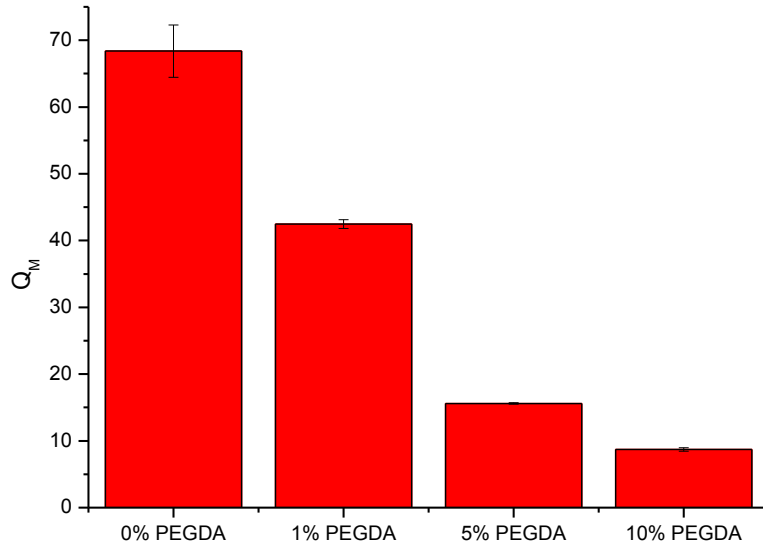
**Figure S1**, FTIR spectra of hyaluronic acid and hyaluronic acid methacrylate (HAMA), with appearance of peak at 1455 cm<sup>-1</sup> giving evidence of methacrylation



**Figure S2** <sup>1</sup>H NMR spectrum of Hyaluronic Acid Methacrylate (HAMA). Resonances at 5.6 and 6.2 ppm verified the presence of methylene protons.

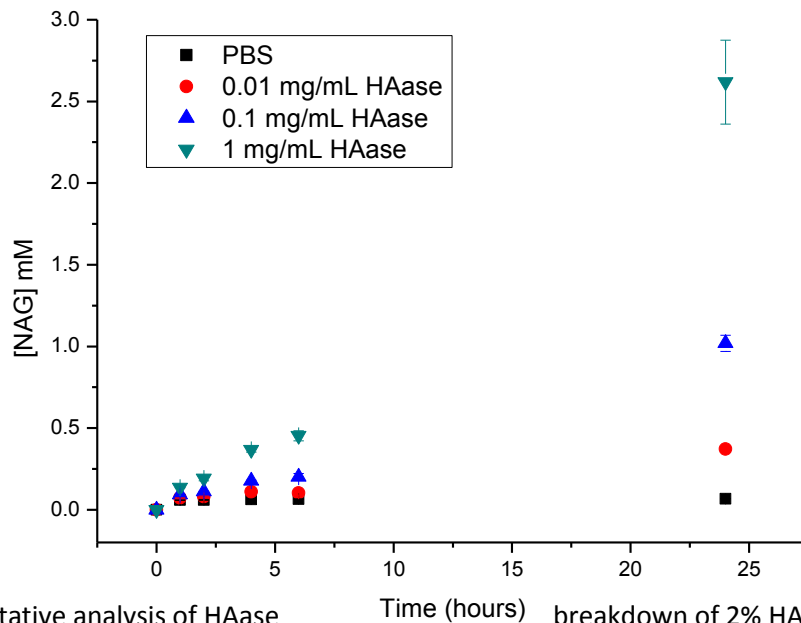


**Figure S3** Carbazole assay reaction mechanism

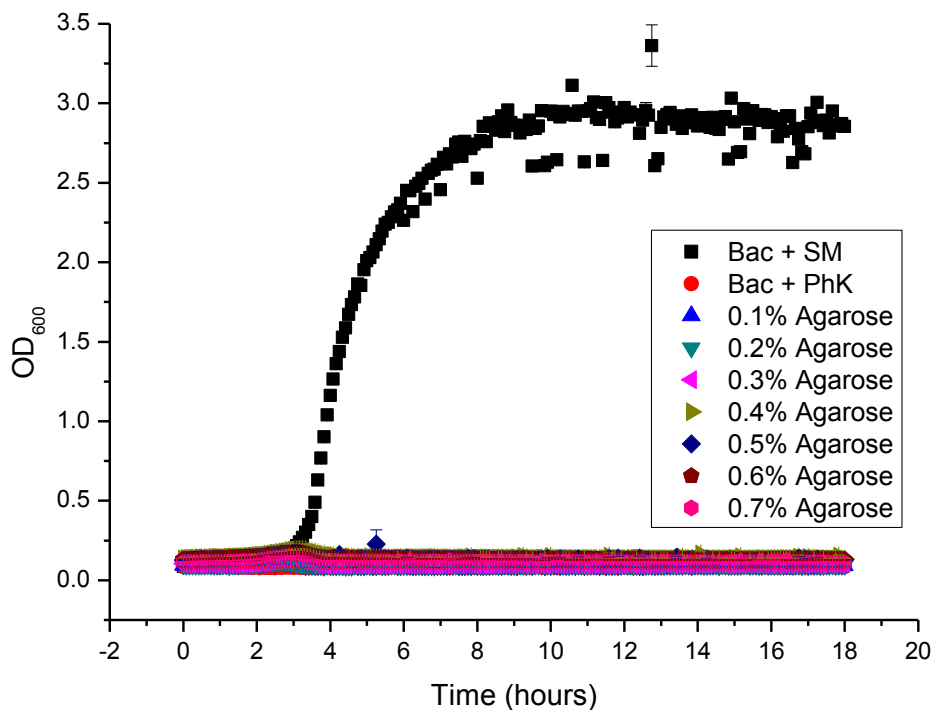


	<u>Swelling ratio, <math>Q_M</math></u>	<u>Volumetric Swelling Ratio, <math>Q_V</math></u>	<u>Florey Parameter, <math>\chi</math></u>	<u>Specific volume of dry polymer, <math>v(\text{cm}^3/\text{g})</math></u>	<u>Average molecular weight between XL, <math>M_c(\text{g/mol})</math></u>	<u>Effective Crosslink Density, <math>V_e(\text{mol}/\text{cm}^3)</math></u>	<u>Mesh Size, <math>\xi(\text{nm})</math></u>
<b>0%PEGDA</b>	<b>68.4±3.9</b>	<b>83.8</b>	<b>0.473</b>	<b>0.575</b>	<b>7.45E+07</b>	<b>1.65E-08</b>	<b>6604</b>
<b>1%PEGDA</b>	<b>42.5±0.7</b>	<b>52.0</b>	<b>0.473</b>	<b>0.575</b>	<b>2.26E+07</b>	<b>5.45E-08</b>	<b>3099</b>
<b>5%PEGDA</b>	<b>15.6±0.1</b>	<b>19.0</b>	<b>0.473</b>	<b>0.575</b>	<b>1.81E+06</b>	<b>6.78E-07</b>	<b>627</b>
<b>10%PEGDA</b>	<b>8.7±0.3</b>	<b>10.5</b>	<b>0.473</b>	<b>0.575</b>	<b>4.13E+05</b>	<b>2.98E-06</b>	<b>246</b>

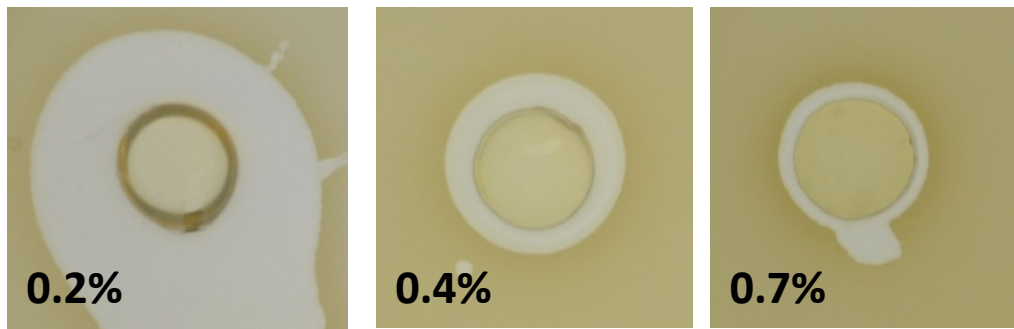
**Figure S4** Swelling ratio  $Q_M$  of HAMA-co-PEG hydrogels



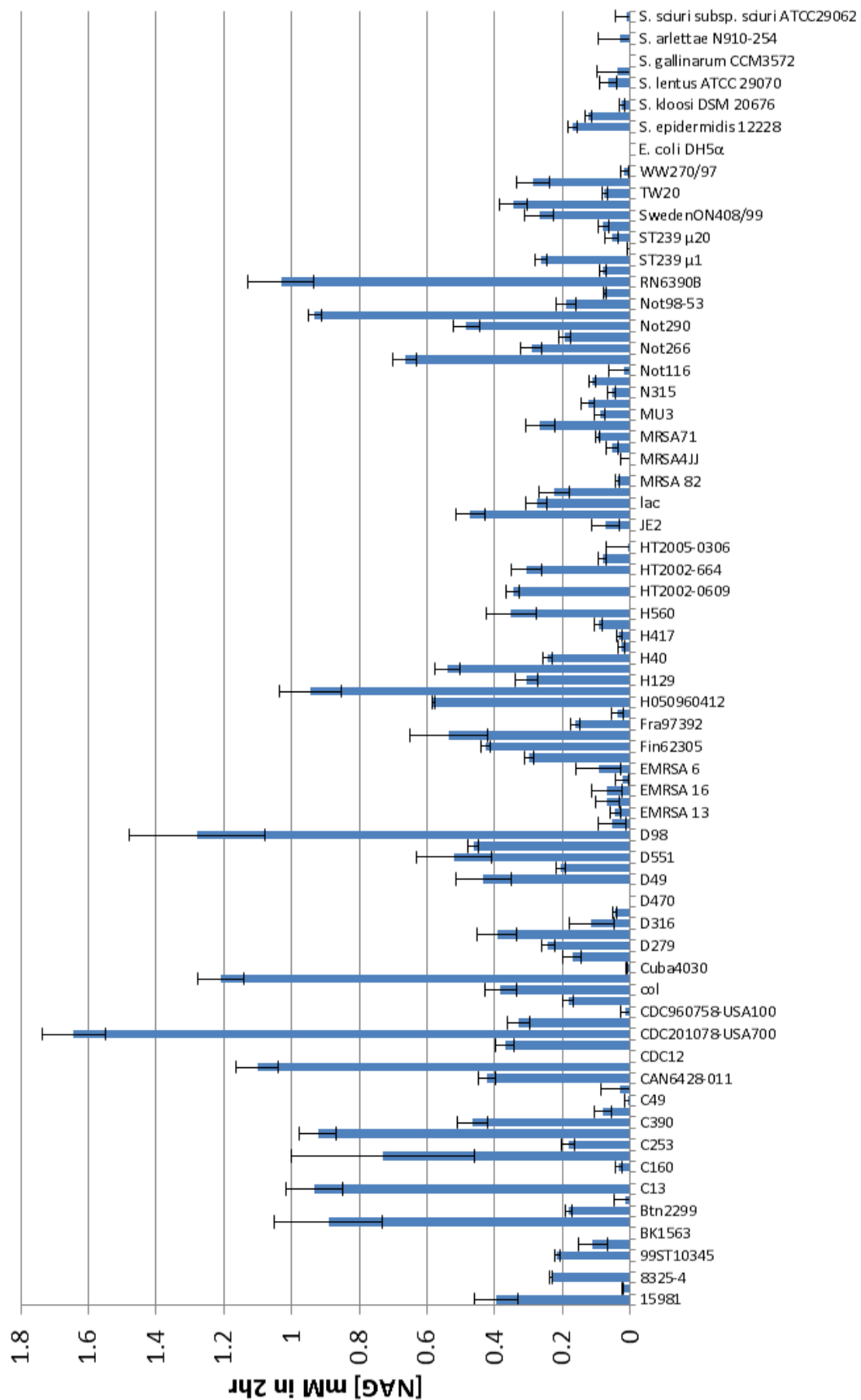
**Figure S5** Quantitative analysis of HAase breakdown of 2% HAMA /1% PEGDA gels, measured via the release of NAG (measured using the Carbazole assay) as a function of incubation time and concentration of hyaluronidase.



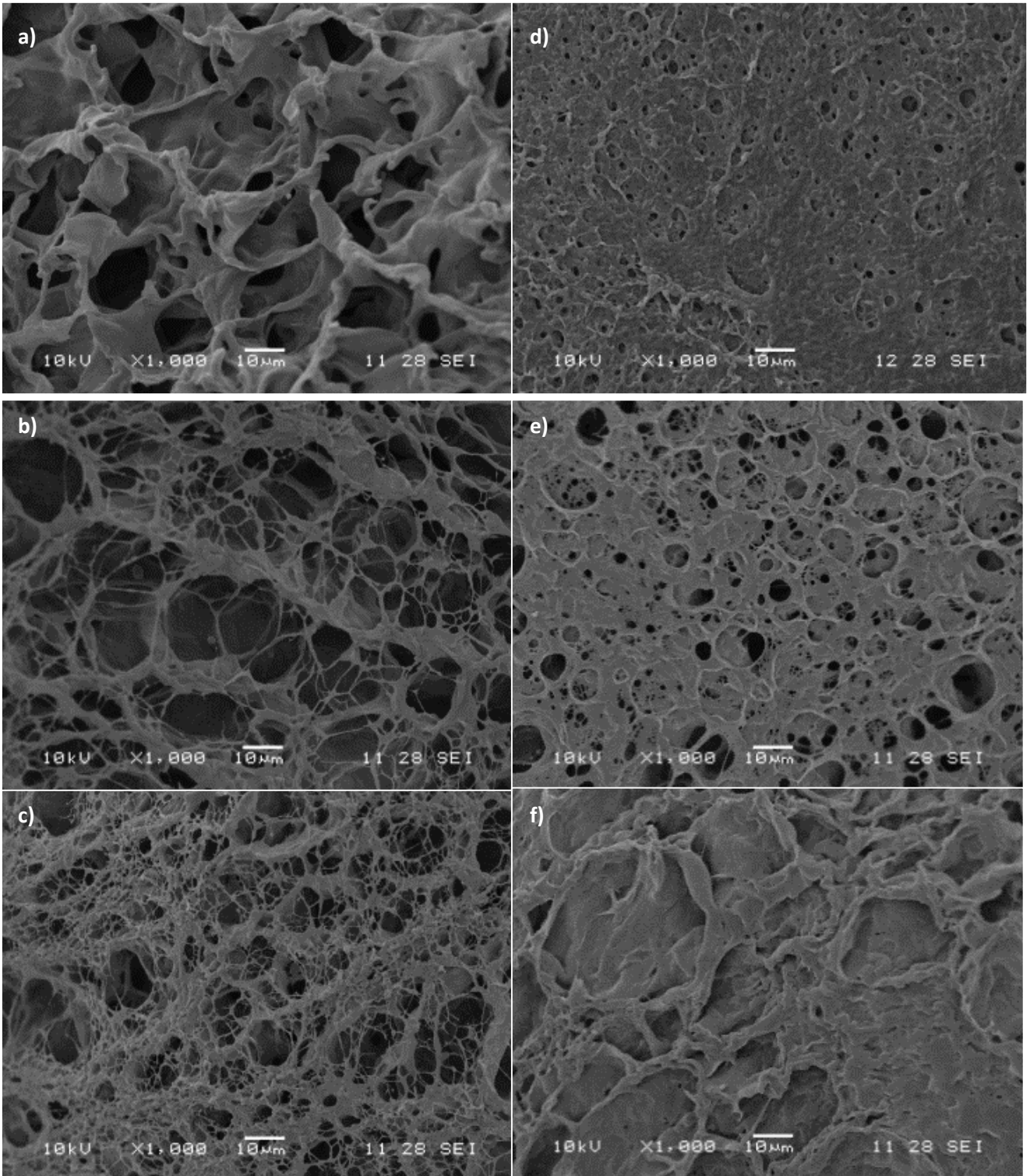
**Figure S6** Phage K in agarose – killing of *S. aureus* H560: Complete suppression of growth / killing seen at all agarose concentrations.



**Figure S7** Bacteriophage K lysis of *S. aureus* H560 bacterial lawns on agar in 0.2%, 0.4% and 0.7% agarose



**Figure S8** Screen of 116 *Staphylococcus* sp. and strains for HAase activity. Concentrations of NAG > 0.4 mM are considered very high, under 0.01 mM low and between these values intermediate, but sufficient to trigger phage release.



**Figure S9** SEM images of HAMA hydrogels after 2h incubation with *S. aureus* supernatant. HAase positive strains: a) RN6390B, b) H560, c) lac. HAase negative strains: d) hys-, e) Mμ2, and f) TSB growth medium.