Electronic Supplementary Material (ESI) for Analyst.

Quantification of a bacterial secondary metabolite by SERS combined with SLM extraction for bioprocess monitoring

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Due to the morphology of the nanostructured SERS surface, based on silicon leaning nanopillars, the percentage of organic solvent and the amount of salts in the sample could influence SERS performance. Therefore, the effect of different EtOH percentages in the standard samples used for quantification was evaluated. 5 µM to 250 µM pHCA were spiked in EtOH/PB with 50% and 90% EtOH v/v. 2 µL droplets were poured on SERS chips and the intensity at 1169 cm^{-1} was analyzed according to the methods described in the Experimental section. As shown in Fig. S1, the two curves are comparable with similar intensity at the same concentration.

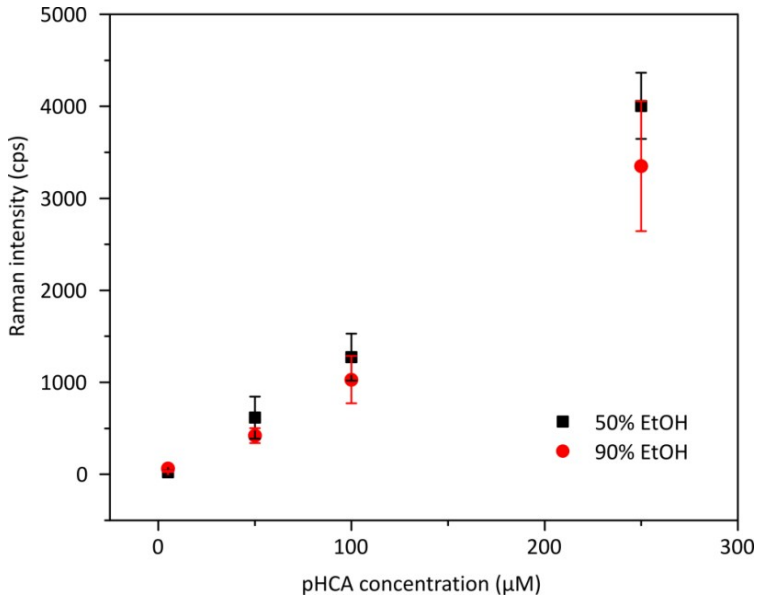

Fig. S1: Raman intensity at 1169 cm^{-1} versus pHCA concentration. Each point was collected at least on duplicate chips, with error bars representing the overall standard deviation.

In order to confirm Tyr exclusion by SLM extraction, we performed a control experiment where we measured the SERS signal in acceptor buffer for pHCA and Tyr separately and together, assuming that Tyr is enriched with the same EF as pHCA. We spiked the acceptor buffer with 500 µM pHCA and 1.5 mM Tyr, both separately and mixed together, diluted the samples 10-fold with EtOH and compared their SERS spectra. In order to better highlight Tyr and pHCA peaks, we subtracted the buffer signal from all the spectra. As it can be seen in Fig. S2, Tyr peaks significantly overlap with pHCA signal between 1150 cm^{-1} and 1300 cm^{-1}. However, when we perform SLM extraction on the samples, we obtain a distinct pHCA signal without Tyr interference, as shown in Fig. 3 c).

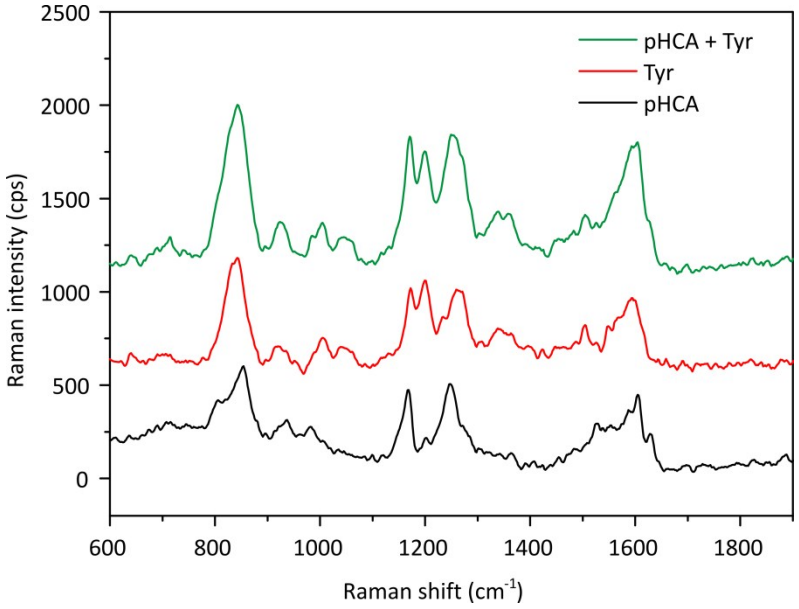

Fig. S2: Comparison between SERS spectra of acceptor buffer spiked with 500 µM pHCA, 1.5 mM Tyr, and both, diluted 10-fold with EtOH. Each spectrum was obtained as the average of 48 points, collected on the surface of a chip wetted with a 2 µL droplet.

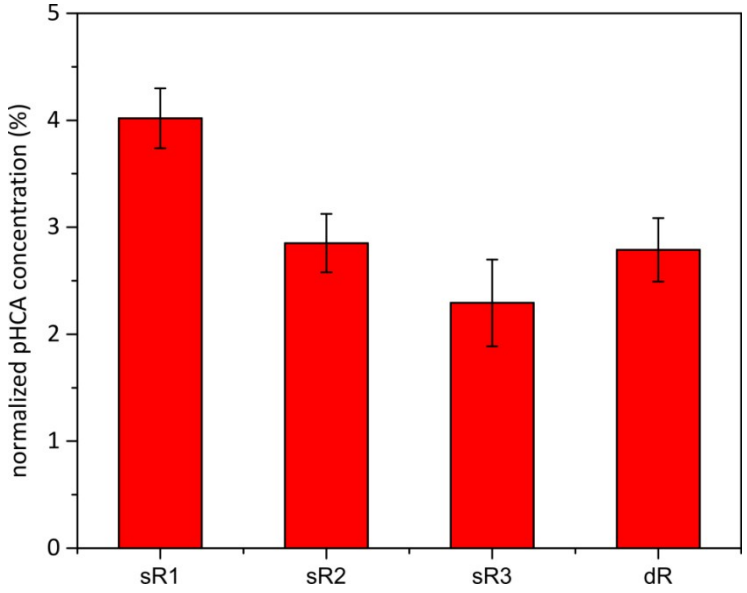

Fig. S3: Effect of different rinsing techniques on memory effect. pHCA concentration in the acceptor after repeated static rinsing (sR1, sR2 and sR3), and after one dynamic rinsing (dR).