Supporting Information

Comparison of voltammetric methods used in the interrogation of electrochemical aptamer-based sensors

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DNA sequences

Table S1. The sequences of the employed aptamers.

<table>
<thead>
<tr>
<th>Target</th>
<th>Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vancomycin</td>
<td>5'-/5ThioMC6-D/CGA GGG TAC CGC AAT AGT ACT TAT TGT TCG CCT ATT GTG GGT CGG/3MB/-3'*</td>
</tr>
<tr>
<td>Doxorubicin</td>
<td>5'-/HS-(CH(_2))(_6)/ACC ATC TGT GTA AGG GGT AAG GGG TGG T/(CH(_2))(_7)-NH-MB/-3'*</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>5'-/5ThioMC6-D/CGA CCG CGT TTC CCA AGA AAG CAA GTA TTG GTT GGT CG/3MB/-3'</td>
</tr>
</tbody>
</table>

*MB = Methylene Blue

Parameters for frequency-gain maps

Table S2. The parameters used in creating frequency-gain maps for each method.

<table>
<thead>
<tr>
<th>Method</th>
<th>Amplitudes (mV)</th>
<th>Increment (mV)</th>
<th>Frequencies (Hz)</th>
<th>Pulse Period (s)</th>
<th>Pulse Width (s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SWV</td>
<td>10, 20, 25, 30, 40</td>
<td>3</td>
<td>8, 10, 15, 20, 25, 30</td>
<td>[0.01, 0.025, 0.05, 0.1, 0.25, 0.5, 0.75]</td>
<td>0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05, 0.125, 0.25</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1</td>
<td>40, 50, 60, 70, 80, 90, 100, 150, 200, 250, 300, 400, 500, 600, 700, 800, 900, 1000</td>
<td>0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05, 0.125, 0.25</td>
<td></td>
</tr>
<tr>
<td>ACV</td>
<td>10, 20, 25, 30, 40</td>
<td>6</td>
<td>8, 10, 15, 20, 25, 30, 40, 50, 60, 70, 80, 90, 100, 150, 200, 250, 300, 400, 500, 600, 700, 800, 900, 1000</td>
<td>0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05, 0.125, 0.25</td>
<td></td>
</tr>
<tr>
<td>DPV</td>
<td>25</td>
<td>3</td>
<td>0.25</td>
<td></td>
<td>0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05, 0.125</td>
</tr>
<tr>
<td></td>
<td>10, 20, 25, 30, 40</td>
<td></td>
<td></td>
<td></td>
<td>0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05, 0.125</td>
</tr>
</tbody>
</table>
Comparison of raw currents recorded with SWV, DPV and ACV

Figure S1. Shown is a comparison of SWV, DPV and ACV raw currents in buffer in the absence of target. Each was measured using the optimal (highest gain) signal-on parameters: 300 Hz for SWV, 0.025 s and 0.0025 s for the pulse period and pulse width, respectively, for DPV, and 50 Hz for ACV.

Titration curves collected in PBS using signal-on and signal-off parameters

Figure S2. Shown are titration data recorded at separate signal-on (darker colors) and signal-off (lighter colors) parameters for each method, collected in 37°C PBS. For SWV the signal-on and signal-off frequencies are 300 and 20 Hz (A), for DPV the pulse period and pulse width are 0.025 s and 0.0025 s and 0.1 s and 0.025 s for the two cases, respectively (B), and for ACV the frequencies are 50 and 5 Hz (C). Error bars are standard deviations obtained from four independent electrodes.
Dependence of signal gain on DPV amplitude

![Dependence of signal gain on DPV amplitude](image)

**Figure S3.** DPV signal gain is strongly dependent on the pulse width and only slightly dependent on the amplitude used. These measurements were done with a pulse period of 0.25 s and pulse widths 0.001, 0.0025, 0.005, 0.0125, 0.025, 0.05 and 0.125 s in 37°C PBS. The error bars represent standard deviations obtained from four independent electrodes.

**Noise levels**

We calculated noise levels associated with each voltammetric method with a custom-made Matlab script using cubic splines (Matlab's csaps function) to obtain a “noise-free” estimate of the noisy measurements. Cubic splines are a widely used way to fit curves to noisy data without employing any specific model beyond the assumption that the fitted curve is “sufficiently smooth.” The degree of smoothness that is sufficient can be chosen by the user for the specific application (argument p for csaps function). The code employed here is available at [https://github.com/PlaxcoLab/comp_voltamm_EAB.git](https://github.com/PlaxcoLab/comp_voltamm_EAB.git).

**Calibration curves in whole blood**

The titration curves collected in whole blood were measured as those collected in buffer. Specifically, we employed the optimal signal-on and signal-off parameters and sequentially injecting more target into the sample solution. The amplitudes used were 25 mV for SWV and DPV and 20 mV for ACV. The KDM values were then calculated from the signal-on and signal-off peak currents at each concentration and the data was fitted with a Langmuir isotherm using
OriginPro 2021b software. Based on these fits, the equations used for retrieving concentrations from the drift data were $c = (KDM \times 150.8)/(0.83 - KDM)$ for SWV, $c = (KDM \times 164.8)/(0.90 - KDM)$ for DPV and $c = (KDM \times 66.6)/(0.49 - KDM)$ for ACV.

**Figure S4.** Shown are vancomycin calibration curves collected at 37°C in whole blood. For SWV, the signal-on and signal-off parameters were 300 and 20 Hz. For DPV the pulse period and pulse width were 0.025 s and 0.0025 s and 0.1 s and 0.025 s, respectively. And for ACV 50 and 5 Hz. Solid lines show the data fitted with a Langmuir isotherm. Error bars are standard deviations obtained from four, independently fabricated electrodes.