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Pathogen-Imprinted Polymer Film Integrated probe/$Ti_3C_2T_x$ MXenes Electrochemical Sensor for Highly Sensitive Determination of *Listeria Monocytogenes*

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Reagents and apparatus

Ti_3AlC_2 (99%) was purchased by Forsman Technology Company (Beijing); hydrofluoric acid (≥ 40%) was purchased from Aladdin Biotechnology Co., Ltd (Shanghai); Th, $K_3[Fe(CN)_6]$, and $K_4[Fe(CN)_6]$ were purchased from Sigma-Aldrich Co., LLC.; PBS was prepared with Na_2HPO_4, NaH_2PO_4 and KCl. The LM cultivation was performed as follows: LM strain was cultured first in LB liquid medium overnight with continuous shaking at 37°C; then, it was centrifuged and resuspended in PBS (pH 7.4), which was repeated for three times to achieve purified LM suspension. The concentration of LM was confirmed in triplicate via plate counting method, and the serial dilutions from LM were prepared with sterilized PBS. All the electrochemical studies were carried out via a CHI 660E Electrochemical Workstation with a common 3-electrode system: GCE, Ag/AgCl electrode and platinum wire.

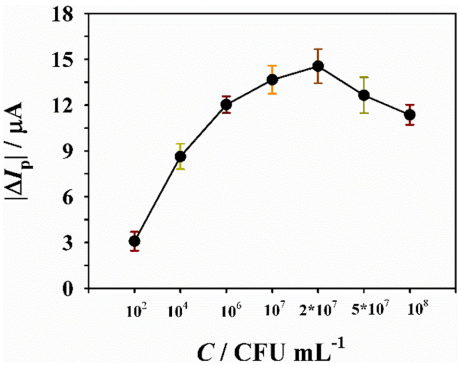

Fig. S1. The influence from the concentration of template LM

From Fig. S1, it's noted that the $|\Delta I_p|$ values increase initially with the increase of LM concentrations from 10^2 to 2×10^7 CFU mL^{-1}, whereas a decrease presented when the LM concentrations increased further. The reason maybe that when the LM template concentration is very low, there are only a few LM cells embedded in the polymer, resulting the generation of a small quantity of imprinted cavities. Conversely, when the template concentrations are too high, the elution of template becomes to be more difficult, still inducing in fewer imprinted sites and poor re-binding ability to the LM cells, hence the template concentration of 2×10^7 CFU mL^{-1} was selected in the imprinting step for preparing PIP/$Ti_3C_2T_x$R/GCE.

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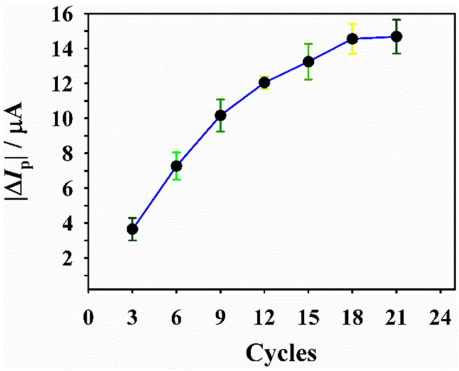

Fig. S2. The influence from the polymerization cycles of Th

Fig. S2 shows the effect from the electropolymerization cycles of Th, it can be found that the $|\Delta I_p|$ values increase with the increase of cycles, and there is almost no obvious increase when the cycles increased to 18, which may be resulted from that the excessive thick film resulted barriers to remove the LM template effectively. Therefore, 18 cycles were used for the electropolymerization process of Th.

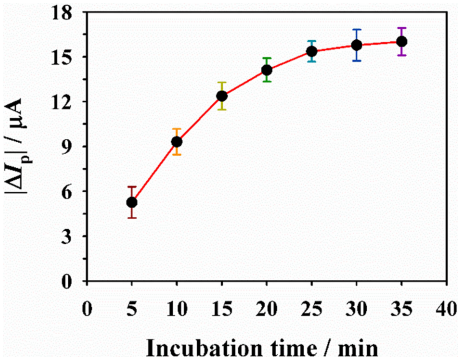

Fig. S3. The influence from the incubation time of PIP/$Ti_3C_2T_x$R/GCE in LM solution

Undoubtedly, the incubation time of PIP/$Ti_3C_2T_x$R/GCE in LM solution is an important parameter. The results from Fig. S3 revealed that the increase of incubation time can effectively increase $|\Delta I_p|$ value which reflect the improvement of analytical performances, while there are only few increases when the incubation time exceed more than 25 min, respectively. Thus, the time of 25 min was selected for the incubation time.

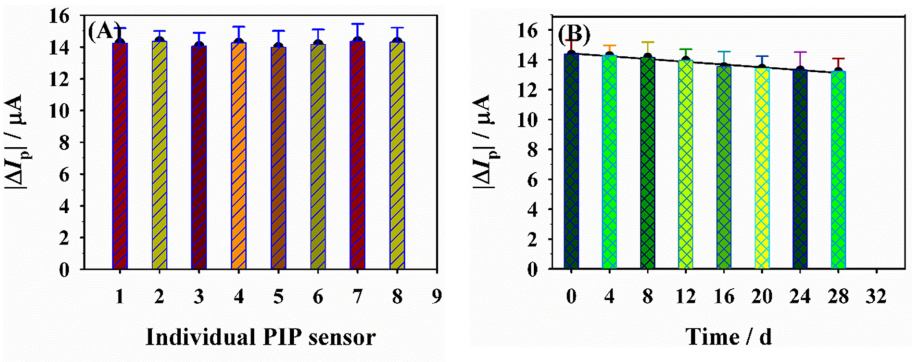

Fig. S4. (a) The reproducibility and (b) stability of the as-prepared PIP/$Ti_3C_2T_x$R/GCE