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Supplementary Information

Transparent crystalline cubic SiC-on-glass electrodes enable simultaneous electrochemistry and optical microscopy

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1. Growth process of the SiC films

The 3C-SiC films were deposited on a Si(100) wafers using a hot-wall LPCVD reactor at 1250°C, Fig. S1. Silane (SiH4) and propylene (C3H6) were utilized as the precursors in our alternating supply epitaxy (ASE) approach to achieve single crystalline SiC films. To form highly doped n-type 3C-SiC, we introduced NH₃ as the dopants during the film deposition. Particularly, the deposition process started with a carbonization step followed by repeating the cycles below to achieve 700-nm thick SiC films:

- (i) Supply of 2sccm SiH₄ for 10 s
- (ii) Pump out for 30 s
- (iii) Supply NH₃ for 10-20 s (in situ doping)
- (iv) Supply 1.5 sccm C₃H₆
- (v) Pump out for 30 s

Each growing cycle form approximately 0.6 nm, and the thickness of the film was controlled by the number of cycles.

2. Fabrication process of SiC-on-glass electrodes

Prior to the bonding process, the SiC/Si wafer and the borofloat glass wafer were cleaned with Pyranha and rinsed with DI water. The surface of the two wafers was then activated using O₂ plasma. Next, the two wafers were brought into contact, under an applied force of 2500 N. Anode electrode was applied to the Si side of the SiC/Si wafer while the cathode was connected to glass, enabling the bonding process. The bonded wafer was then immersed into KOH (30%wt) at 80°C to completely remove the Si layer, resulting in the SiC-on-glass platform. Subsequently, the SiC-on-glass wafer was cleaned using the standard RCA procedure to remove the residual KOH etching contaminations.

A 300nm thick Al film was deposited on the bonded SiC film and then wet-etched to form contact areas. Polyimide was then spin-coated, plasma etched, and then cured at 200°C for 1 hour to define the functional surface area of working electrode (i.e. SiC). Finally, the SiC-onglass wafer was diced to form smaller electrodes for subsequent experiments.

3. Mechanical strength of SiC and glass bonding

To investigate the mechanical stability of the platform we measure adhesion force between the SiC film and the glass substrate using a tensile pulling machine Shimadzu TMAG-X. We diced the bonded Si/SiC/glass wafer into $10\text{mm} \times 10\text{mm}$ chips and then mounted onto the copper jigs using epoxy, Figure S2. We applied tensile stress to the samples until reaching the fracture point. The results showed that the adhesion force of all samples was above 20 MPa, indicating a strong mechanical bonding between the SiC film and the glass substrate.

4. Electrochemical measurement

4.1 Materials

Unless otherwise stated, the reagents and chemicals used for the conducting experiments were of analytical grade. Reagent grade hexaammineruthenium(III) chloride (RuHex), methylene

blue (MB), phosphate buffer saline (PBS) tablet (0.01M phosphate buffer, 0.0027M potassium chloride and 0.137M sodium chloride, pH 7.4 at 25°C) were obtained from Sigma-Aldrich (Australia). Tris was purchased from VWR Life science (Australia).

4.2 Electrochemical measurement

All electrochemical measurements were performed with a CHI650 electrochemical workstation (CH Instrument, USA). Cyclic voltammetry (CV) experiments were done in a single-compartment cell. A conventional three-electrode system, comprising a SiC working electrode, a platinum auxiliary electrode, and an Ag/AgCl₃ 1.0 M NaCl reference electrode (CH Instrument, Inc. USA), was used for the measurement of electrocatalytic activity.

Figure S3 demonstrates the excellent repeatability of the CV after numerous scan cycles. Figures S4, S5, S6, S7 and S8 show the CV of SiC-on-glass in another commonly used electrolyte (methylene blue).

5. Cell culture and stimulation methodology

5.1 Cell culture procedure

For sterilization, SiC-on-glass sampled were cleaned with 80% ethanol and then washed with 1X PBS. This was followed by 1-hour UV exposure. Subsequently, the samples were incubated with fresh media at 37°C and 5% CO2 for 1 hour before the cell seeding. The 25 cm² culture flasks, each containing 1×10^6 viable cells of human Mammary Fibroblasts (HMF) were cultured in DMEM/F12 containing 10% FBS and 1% penicillin–streptomycin. The cells were maintained in the standard incubator at 37°C and 5% CO2. For seeding the device, cells were harvested from 80% confluent using TrypLE Express (Thermo Fisher Scientific) and resuspended in the culture medium. Aliquots with an optimised seeding density of 2.5×10^4 cells/500 μL and were used to seed the well of the device. Finally, the SiC-on-glass plate was incubated at 37°C and 5% CO2 for 18 hours. Further, to ensure the confirm the cytocompatibility, adherence and growth of the cells onto the SiC-glass plate was observed and imaged at 24, 48, and 72 hours using Nikon EclipseTs2 phase contrast microscope with 10 X magnification.

5.2 Cell lysis at applied voltage of 10V

A PMMA well was attached on the SiC chips for cell culture. To avoid the leakage of culturing media, UV gel (Norland Optical Adhesive 81) was used as the adhesive layer between the PMMA well and the SiC-on-glass chip. The Al electrode pads on the SiC chips was connected to an external Copper/PCB using wire bonding, as shown in Figure S9.

Experimental results also evidenced that increasing the voltage up to 10V led to a complete cell lysis near the vicinity of the SiC heater, which was visualized under a fluorescent microscope. Figure S10 shows the florescent image of the death cells around the SIC heaters (stained with PI).

Supporting figures

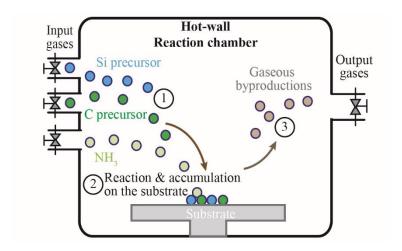


Figure S1. Schematic sketch of the LPCVD process

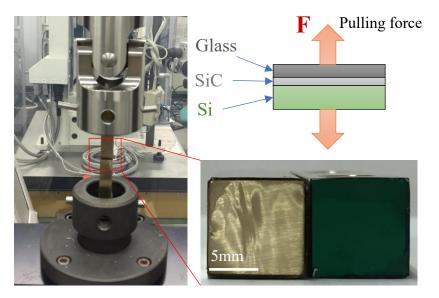


Figure S2. Pulling test showing excellent mechanical property of SiC/glass bonding. Bottom right: The chip failed within the glass bulk-layer, but not at the SiC/glass interface.

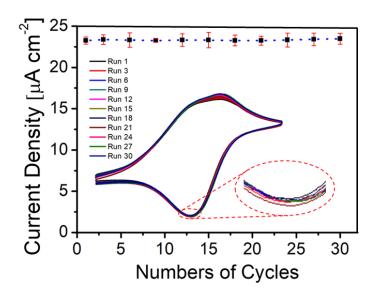


Figure S3. Cathodic peak current versus number of CV cycle (50 μ M RuHex, 0.1 M PBS, pH 7.4); *inset:* corresponding CVs from cyclic run 1 to 30.

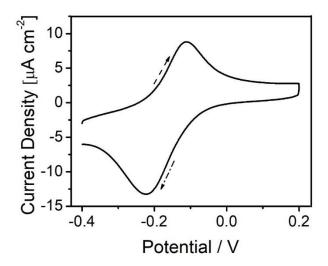


Figure S4. Cyclic voltammograms of single crystal SiC electrode in 5 μ M (in 0.1 M PBS) at the scan rate of 0.1 Vs⁻¹. Two characteristics peaks at 223 mV and 108 mV respectively for cathodic and anidic potential with potential difference 31 mV, revealing the two-electron redox process of MB.

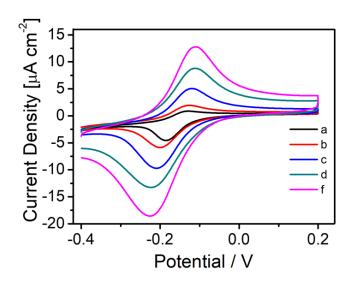


Figure S5. Cyclic voltammograms obtained at SiC electrode at different scan rate ranging from 0.01 to 1.5 Vs⁻¹ (5 μ M MB, 0.1 M PBS, pH 7.4).

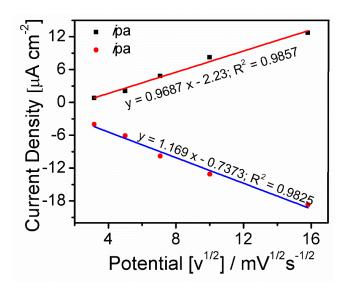


Figure S6. Corresponding curves (obtained at SiC electrode) for i_{pa} and i_{pc} (anodic and cathodic peak current density) as a function of square root of potential $(v^{1/2})$ ranging from 0.01 to 1.5 Vs⁻¹.

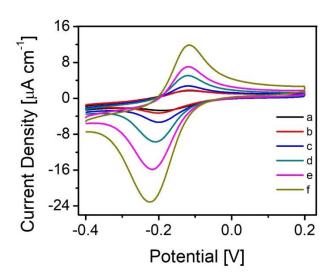


Figure S7. Cyclic voltammograms of SiC electrode upon successive addition of MB (a-0.5, b-1, c-2, d-5, e-10 and f-20 μ M) to the 0.01M PBS (pH-7, scan rate = 50mVs-1).

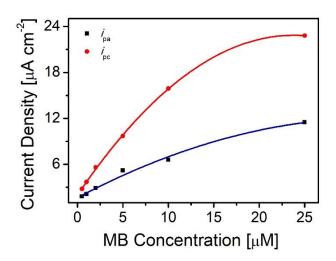


Figure S8. Anodic (blue) and cathodic (red) peak current density at SiC electrode upon different concentration of MB ranging from 0.1 to 25 μ M (0.1 M PBS, pH 7.4, scan rate 0.1 Vs⁻¹). It has clearly been shown that the current density reached a plateau after 10 μ M of MB.

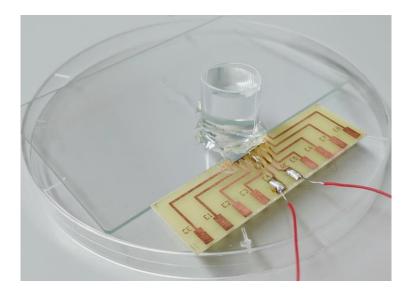


Figure S9. Photographs of a test chip for electrical stimulation using SiC electrodes.

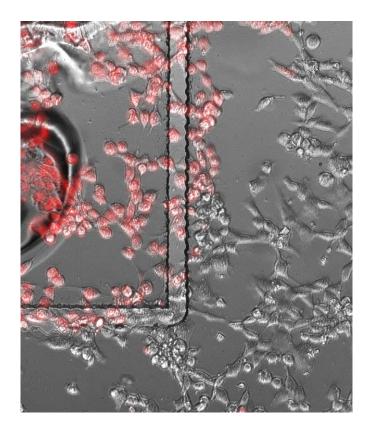


Figure S10. Cell thermal lysis at an applied voltage of 10V.