1 **Supplementary Information**

2 **Vapor grown carbon fiber combined with polyaniline and gold nanoparticles in**

3 **composite bielectrodes and its application in glucose fuel cells**

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6 **Contents**

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16   I.   Procedure of enzyme activity measurement

17   GOx activity of the synthesized composite was determined by measuring the

18   absorbance of upper solution of the composite matrix from centrifugation after

19   immobilization using Glucose Oxidase Activity Assay Kit MAK 097 purchasd from

20   Sigma-Aldrich. In detail, 2 mg PANI/VGCF (or AuNPs/VGCF) was dissolved in 500 μL

21   DI water followed by adding 1 mg GOx while shaking at 200 rpm for 1 h at 0 ℃. 1 μL

22   GA was added to the mixture which was shaken continuously overnight. Then the mixture

23   was centrifuged gently. Sample preparation was finished when upper solution was

24   obtained. H_2O_2 standards were prepared first with 0 (blank), 1, 2, 3, 4, and 5 nmole per

25   tube standards in 1 mL buffer. Dilute samples and standards were measured by

26   colorimetric method. First, mixtures including GOx Assay Buffer (36 μL), GOx

27   Developer (2 μL), Fluorescent Peroxidase Substrate (2 μL), GOx Substrate (10 μL) were

28   added to the samples and standards. After 5 minutes, the initial measurement ($T_{initial}$) was

29   taken. For colorimetric assays, measurements were taken every minute at the absorbance

30   of 570 nm (A_{570}) $_{initial}$ using a UV spectrometer (UV-1800, Shimadzu, Kyoto, Japan). The

31   final measurement (A_{570}) $_{final}$ for calculating the enzyme activity would be the penultimate

32   reading or the value before the most active sample was near or exceeds the end of the

33   linear range of the standard curve. The time of the penultimate reading was T_{final}.

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35   II.   Calculation of GOx immobilized on PANI/VGCF (or AuNPs/VGCF) composite

36   The amount of GOx immobilized on PANI-VGCF (or AuNPs/VGCF) composite

37   was determined by subtracting the amount of GOx in the upper solution of composite

38   matrix from centrifugation after immobilization from the total amount. For this purpose,

39   we first calculated the upper solution enzyme activity using the following equation.

$$GOx\ Activity = \frac{B\ \times Sample\ Dilution\ Factor}{(Reaction\ Time)\ \times V}$$

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41   where B was the amount (nmole) of H_2O_2 generated between $T_{initial}$ and T_{final}.

42 Reaction Time (minutes) was the time difference between T_{final} and $T_{initial}$. V was the

43 sample volume (mL) added to the tube. GOx activity was reported as nmole min^{-1} mL^{-1}

44 = milliunit mL^{-1}, where one unit of GOx was defined as the amount of enzyme that

45 generates 1.0 mmole of H_2O_2 per minute at 37 °C. After calculating the enzyme activity,

46 the amount of enzyme immobilized onto PANI/VGCF (or AuNPs/VGCF) composite was

47 obtained by subtracting the amount of GOx in the upper solution from the total amount
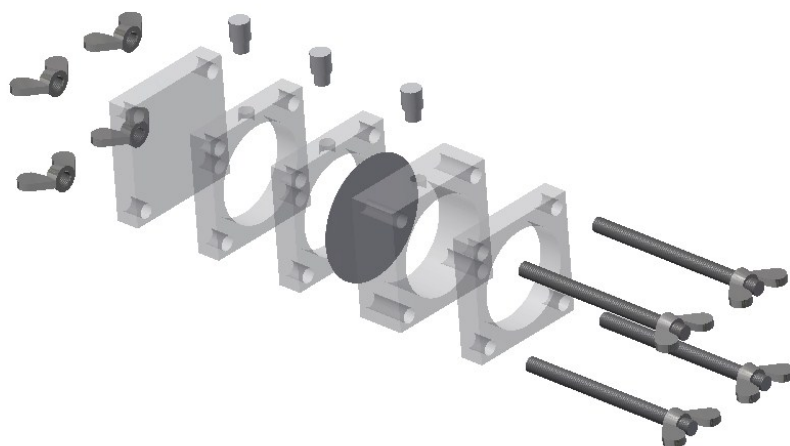
48 of GOx.

III. Figures

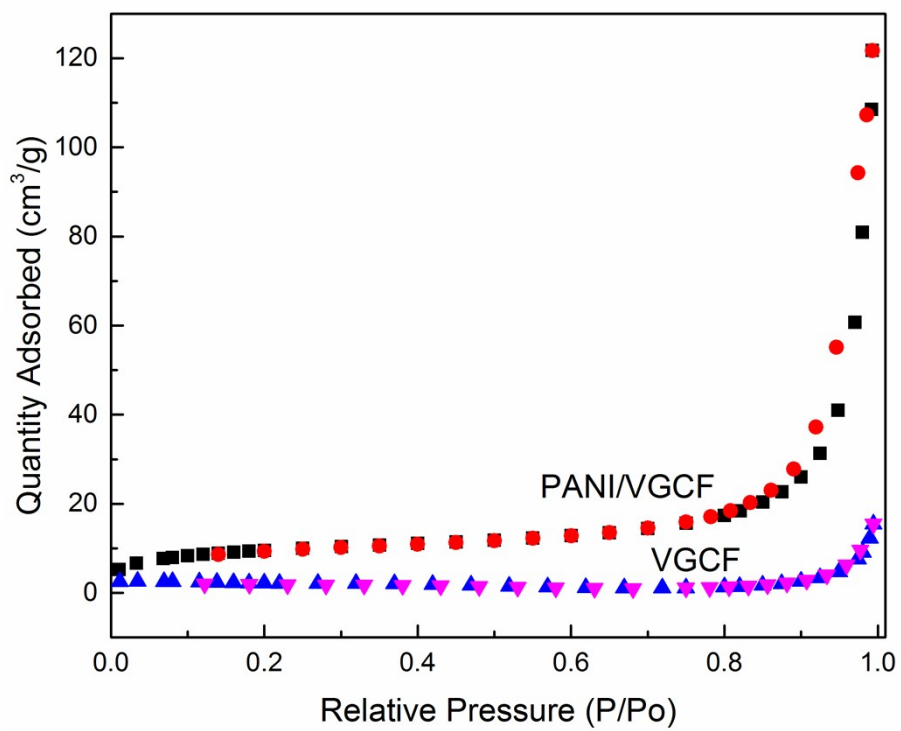**Figure S1.** A schematic of the EBC cell consisting of anode, cathode and Nafion 117 membrane.

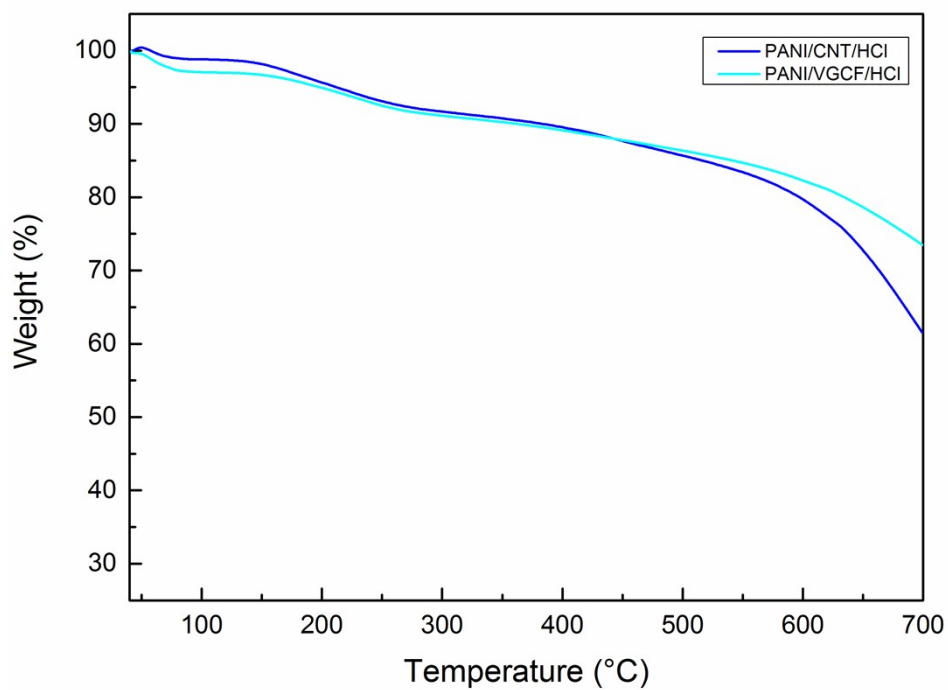**Figure S2.** Isotherm linear plot of HCl doped PANI/VGCF composite and VGCF.

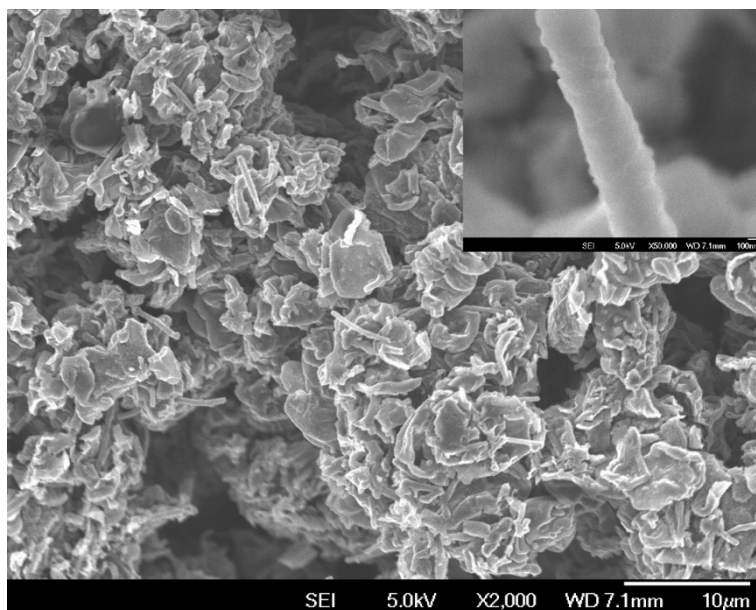**Fig. S3.** TGA of PANI/VGCF composite (cyan) and PANI/CNT composite (blue).

Fig. S4. SEM of PANI/VGCF composite doped with SDS with different magnifications (large: 2000×, inset: 50000×).
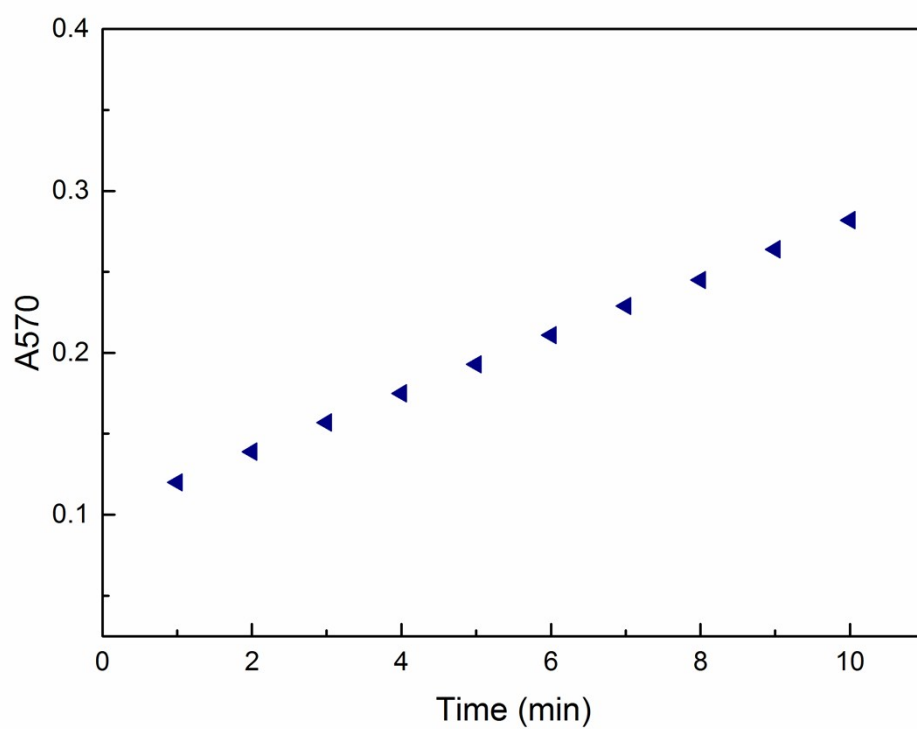
Figure S5. Absorbance vs time (A-T) plot for calculation of enzyme activity of PANI/VGCF composite enzyme matrix.
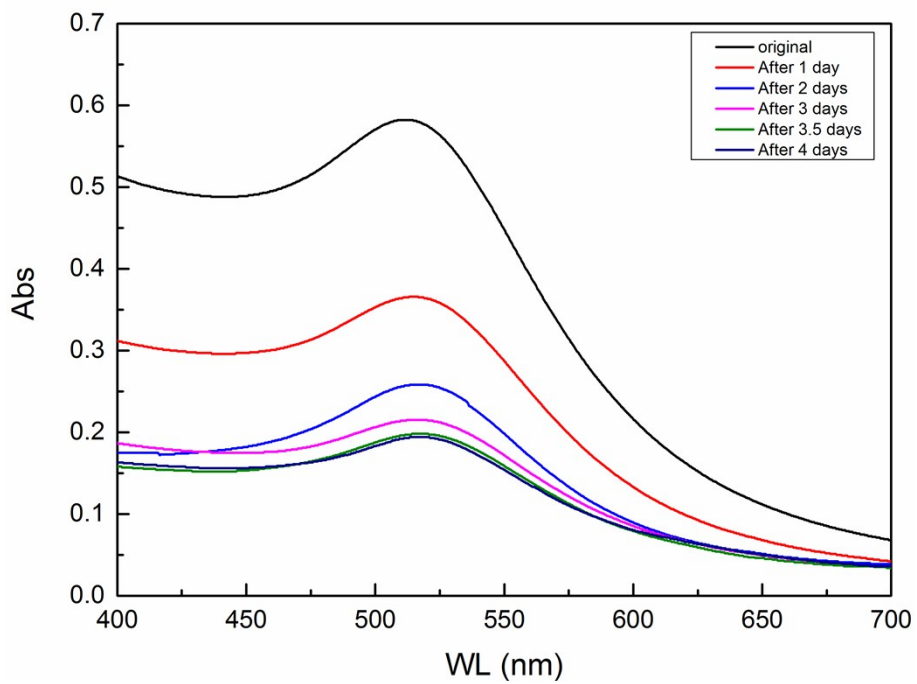
Figure S6. The UV absorbance of residual solution of AuNPs/VGCF with increasing time.
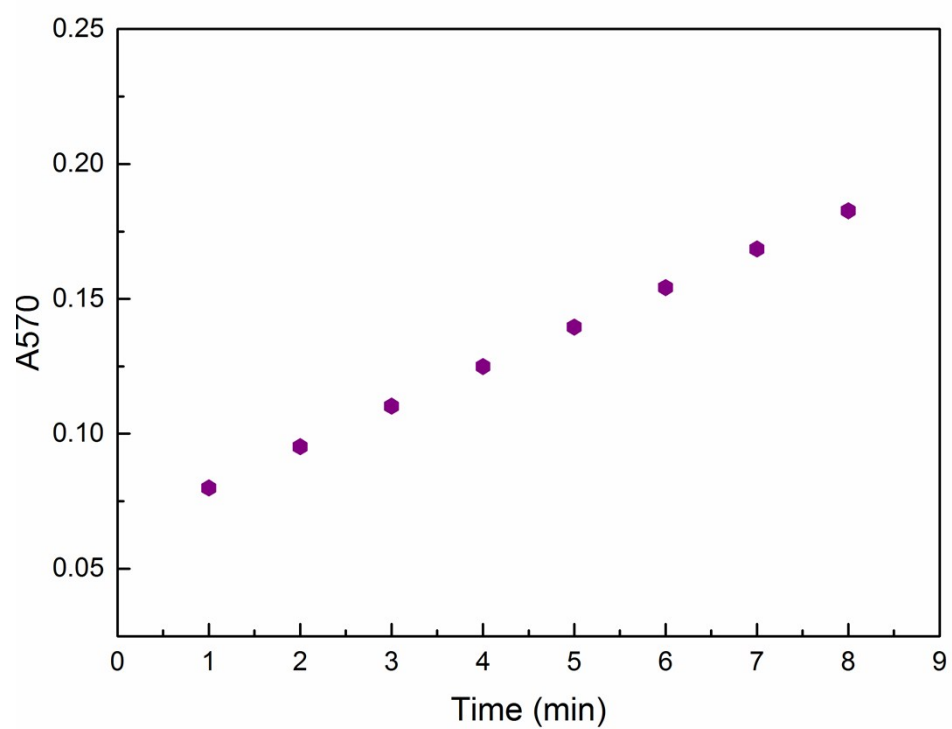
Figure S7. Absorbance vs time (A-T) plot for calculation of enzyme activity of AuNPs/VGCF composite enzyme matrix.