

Electronic Supplementary Information

Non-enzymatic detection of glucose levels in human blood plasma by a graphene oxide-modified organic transistor sensor

Haonan Fan,^a Yui Sasaki,^a Qi Zhou,^a Wei Tang,^a Yuta Nishina^{*b,c} and Tsuyoshi Minami^{*a}

^a Institute of Industrial Science, The University of Tokyo, 4-6-1 Komaba, Meguro-ku, Tokyo, 153-8505, Japan.

Corresponding author: tminami@g.ecc.u-tokyo.ac.jp

^b Research Core for Interdisciplinary Sciences, Okayama University, Okayama 700-8530, Japan,

^c Graduate School of Natural Science and Technology, Okayama University, Okayama 700-8530, Japan.

Corresponding author: nisina-y@cc.okayama-u.ac.jp

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References

1. General

Reagents and materials

Commercially available reagents and supplied materials were applied to the device fabrication and the analyte detection without any pre-treatments.

For fabrication of the OFET device: A glass substrate (model: Eagle XG, 2 cm × 2.5 cm) obtained from Corning, Inc. was utilized. An aluminum (Al) wire (1φ) for a gate electrode and a gold particle (Au) for the source and drain electrodes were respectively purchased from Furuuchi Chemical Co., Ltd. and Tanaka Kikinzoku Kogyo Co., Ltd. Tetradecylphosphonic acid (TDPA) obtained from Tokyo Chemical Industry Co., Ltd. was applied as a dielectric layer. Poly{2,5-bis(3-tetradecylthiophen-2-yl)thieno[3,2-*b*]thiophene} (PBTTC-C14) and 1,2-dichlorobenzene for a polymer semiconductive layer were purchased from Merck KGaA. An amorphous fluoropolymer (CYTOP™, model: CTL-809M) was supplied from AGC Co., Ltd. An organic solvent obtained from Kanto Chemical Co. Inc. was 2-propanol.

For fabrication of the extended-gate electrode: A polyethylene naphthalate (PEN) film was supplied from TOYOBO Co., Ltd. 4-Mercaptophenylboronic acid (4-MPBA) as a glucose receptor and tris(2-carboxyethyl)phosphine (TCEP) hydrochloride as a reducing agent were respectively purchased from Merck KGaA. and FUJIFILM Wako Pure Chemical Industries, Ltd. The functionalization of the receptor was carried out in methanol obtained from Kanto Chemical Co. Inc.

For electrical measurements and chemical sensing: A conductive silver paste (model: D-500), potassium chloride (KCl), sodium chloride (NaCl), disodium hydrogenphosphate dodecahydrate, potassium dihydrogenphosphate dihydrate, hydrogen tetrachloroaurate(III) tetrahydrate (HAuCl₄), human serum albumin, immunoglobulin G from human, glucose, fructose, and ascorbic acid were obtained from FUJIFILM Wako Pure Chemical Industries, Ltd. Plasma from human was obtained from Merck KGaA. The analytes obtained from Tokyo Chemical Industry Co., Ltd. were mannose, galactose, citric acid, and lactic acid. An Ag/AgCl electrode as a reference electrode and a Pt wire as a counter electrode were purchased from BAS Inc. Milli-Q water (18.2 MΩ·cm) was applied to prepare all aqueous solutions in this assay.

Apparatuses

Purpose	Apparatus
Vacuum thermal deposition	SVC-700TMSGs, Sanyu Electron Co., Ltd.
Reactive ion etching (RIE)	SAMCO RIE-10NR
UV/O ₃ treatment	ASM4010Z, ASUMI GIKEN Co., Ltd.
Spin-coating of the hydrophobic layer	MIKASA SPINCOATER 1H-D7
Measurements of the OFET characteristics	Semiconductor parameter analyzer 4156B, Agilent
Electrochemical deposition and measurements	SP-300 potentiostat, Biologic
Photoelectron yield spectroscopy (PYS) in air	AC-2, Riken Keiki, Co.
Field-emission scanning electron microscopy (FE-SEM)	Thermo Fisher Scios2
Energy dispersive X-ray spectroscopy (EDX)	Thermo Fisher Scios2
Contact angle measurements	CA-X contact Angle goniometer, Kyowa

	interface science Co., Ltd.
pH Adjustment	Seven Excellence pH meter, Mettler-Toledo, Ltd.
Determination of plasma glucose concentrations	Biochemical analyzer using a dry chemistry system SP-4430, arkray
ξ-Potential measurements	Zeta-potential & particle size analyzer ELSZ-2000, Otsuka Electronics Co., Ltd.

Procedure for preparation of graphene oxide (GO) solution

Graphite (500 mg) was stirred in 95% w/w H₂SO₄ (25 mL). KMnO₄ (1.5 g) was gradually added to the solution while keeping the temperature <30 °C using an ice bath. The mixture was then stirred at 35 °C for 2 h. The resulting mixture was diluted with water (25 mL) under vigorous stirring and cooling so that the temperature did not exceed 50 °C. The suspension was further treated with 30% aq. H₂O₂ (1.25 mL). The resulting suspension was purified by repeated centrifugation with water. After wet-type jet milling, dialysis was performed until dialysate became neutral.

2. Fabrication and operation of the OFET-based sensor

Fabrication of the OFET

A glass substrate was treated with a piranha solution (H₂SO₄:H₂O₂ = 4:1 (v/v)), followed by ultrasonic treatment in ultrapure water. The washed substrate was applied to the thermal deposition of an Al electrode (30 nm in thickness) with a shadow mask for patterning. The surface of the Al gate electrode was activated by an RIE process to form a gate dielectric layer (AlOx). Next, the spin coater apparatus was applied to form a fluorinated polymer layer made of CYTOP™ (CLT-809M in CT-Solv.180, ratio 1:1 (v/v)) on the substrate, and the annealing process at 110 °C for 10 min was followed. After this period, a hydrophobic bank was formed by the RIE treatment along with a shadow mask. Subsequently, the activated AlOx was treated with TDPA in 2-propanol (10 mM) for 15 h at 25 °C. After immersion in the TDPA solution, the substrate was gently rinsed with 2-propanol and dried with N₂ gas. The annealing process at 110 °C for 30 min was carried out in an inert atmosphere glovebox (UNICO, UL-1300A-MSP). Next, the vacuum thermal deposition was applied to the fabrication of source and drain electrodes made of Au (30 nm in thickness). A solution of PBTTT-C14 (0.0125wt%) in 1,2-dichlorobenzene was drop-cast onto the channel region (width: 50 μm, length: 1000 μm). The substrate was baked at 160 °C for 10 min after drop-casting, obtaining a semiconductive layer. Finally, a passivation layer made of CYTOP™ was fully spin-coated on the surface of the substrate and annealed at 110 °C for 10 min.^{S1,S2}

Operation of the OFET

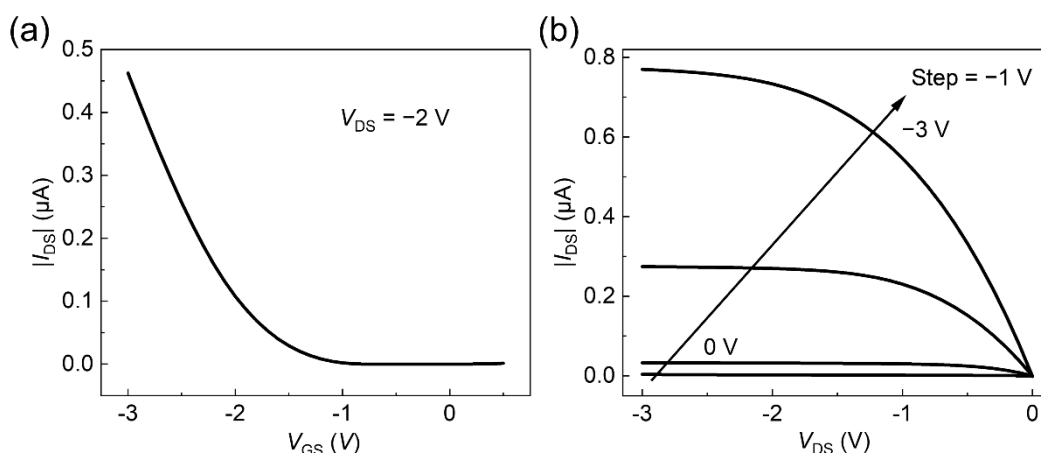


Fig. S1 (a) Transfer curve ($I_{DS} - V_{GS}$) at $V_{DS} = -2$ V and (b) output curves ($I_{DS} - V_{DS}$) at $V_{GS} = 0 - -3$ V. The transistor characteristics of the manufactured OFET were measured under ambient conditions.

3. Fabrication and characterization of the extended-gate Au electrode

Fabrication of the extended-gate Au electrode

An extended-gate Au electrode (15 mm² for sensing area, 100 nm in thickness) was thermally deposited on the PEN film with a shadow mask for patterning. A gold nanostructure (AuNS) was electrochemically deposited on the thermally deposited Au electrode by chronoamperometry with a three-electrode system in a HAuCl₄ solution (100 mM), which was carried out at -0.1 V/s for 1 min. The treated surface of the electrode was rinsed with Milli-Q water. Next, the chronoamperometric method was also applied to the electrochemical deposition of GO on the AuNS electrode in a GO solution (1 mg/mL) at 0.8 V/s for 15 min. After rinsing with Milli-Q water, the electrode was subsequently immersed in a methanol solution containing 4-MPBA (10 mM) and TCEP (50 mM) for 1 h under ambient conditions, and gently rinsed with Milli-Q water and dried with N₂ gas.

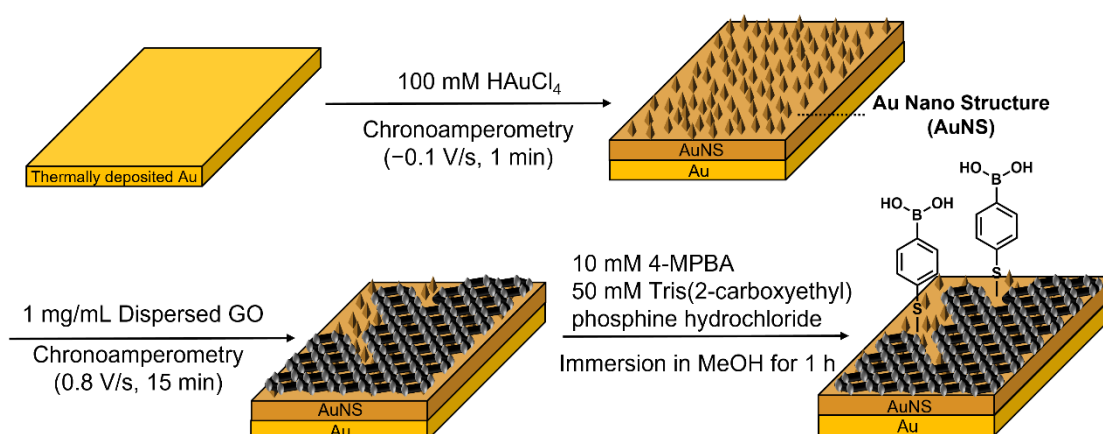


Fig. S2 Schematic illustration of the fabrication procedure for the extended-gate Au electrode.

Liner sweep voltammetry (LSV)

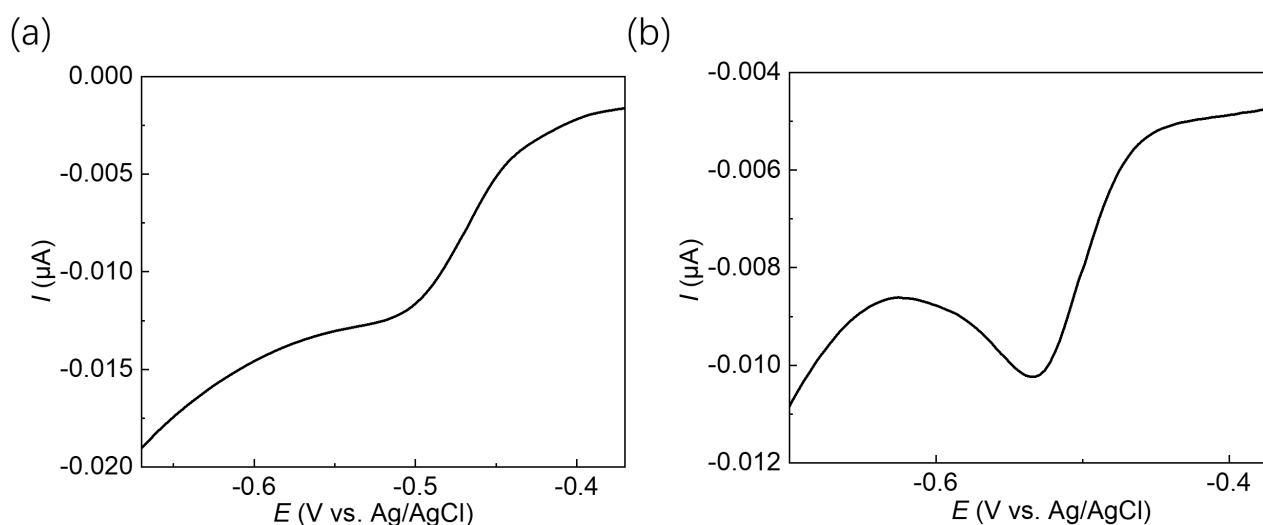


Fig. S3 Determination of the molecular density of 4-MPBA on (a) the GO and AuNS layer ($(8.5 \pm 0.2) \times 10^{-10}$ mol/cm²) and (b) the thermally deposited Au electrode ($(1.13 \pm 0.12) \times 10^{-9}$ mol/cm²). Liner sweep voltammetry in a KOH solution (0.1 M) was performed with four repetitive measurements from 0 V to -1.6 V at 20 mV/s as the scan rate. Faraday's raw with the integration of the peak area was applied to determine the molecular density.

Photoelectron yield spectroscopy (PYS) in air

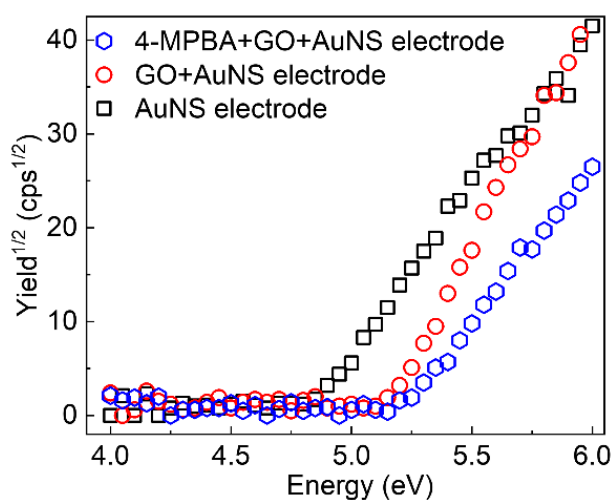


Fig. S4 PYS measurements of the AuNS electrode (black square, 4.89 ± 0.04 eV), the GO+AuNS electrode (red circle, 5.16 ± 0.08 eV), and the GO+AuNS electrode functionalized with 4-MPBA (blue hexagon, 5.18 ± 0.03 eV).

Contact angle measurements

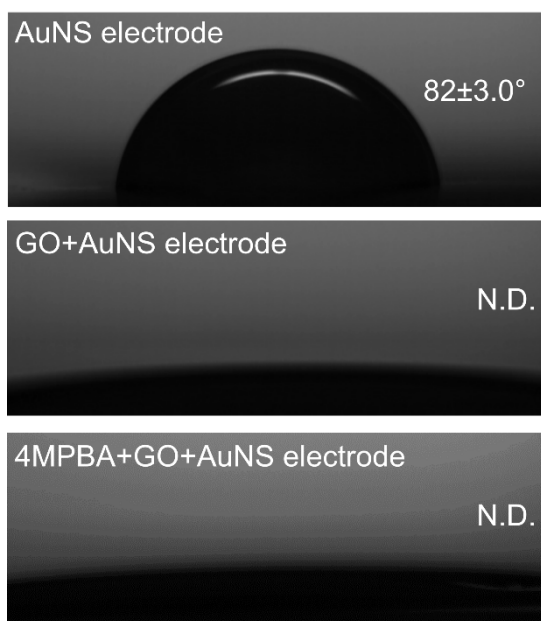


Fig. S5 Contact angles of water droplets on (a) the AuNS electrode, (b) the GO+AuNS electrode, and (c) the GO+AuNS electrode functionalized with 4-MPBA.

4. Investigation of the detectability of the OFET-based sensor

Detection mechanism of the OFET-based sensor

The extended-gate-type OFET sensor consists of the OFET as an operation part and the extended-gate electrode as a sensing part, which is connected through a copper cable.^{S3} The OFET was operated by using a semiconductor parameter analyzer, and the gate voltage (V_{GS}) of the OFET-based sensor was applied through a reference electrode (Ag/AgCl). The transistor characteristic of the OFET (*i.e.*, drain current (I_{DS}) and threshold voltage (V_{TH})) is defined by the following equation (1):

$$I_{DS} = \frac{WC}{2L} \mu (V_{GS} - V_{TH})^2 \quad (1)$$

W and L indicate the channel width and length, respectively; μ means the field-effect mobility; C represents the capacitance of the gate dielectric of the OFET.^{S4} Moreover, a correlation among V_{TH} , C , and the charge density (ΔQ) is described as equation (2).

$$\Delta V_{TH} = \frac{\Delta Q}{C} \quad (2)$$

The changes in surface potential of the extended-gate electrode functionalized with GO and 4-MPBA are induced upon adding target analytes, followed by the changes in the conductance of the OFET. Therefore, the concentration-dependent changes in V_{TH} are caused by the surface changes in the extended-gate electrode accompanied with capturing target saccharides on the 4-MPBA-based SAM.

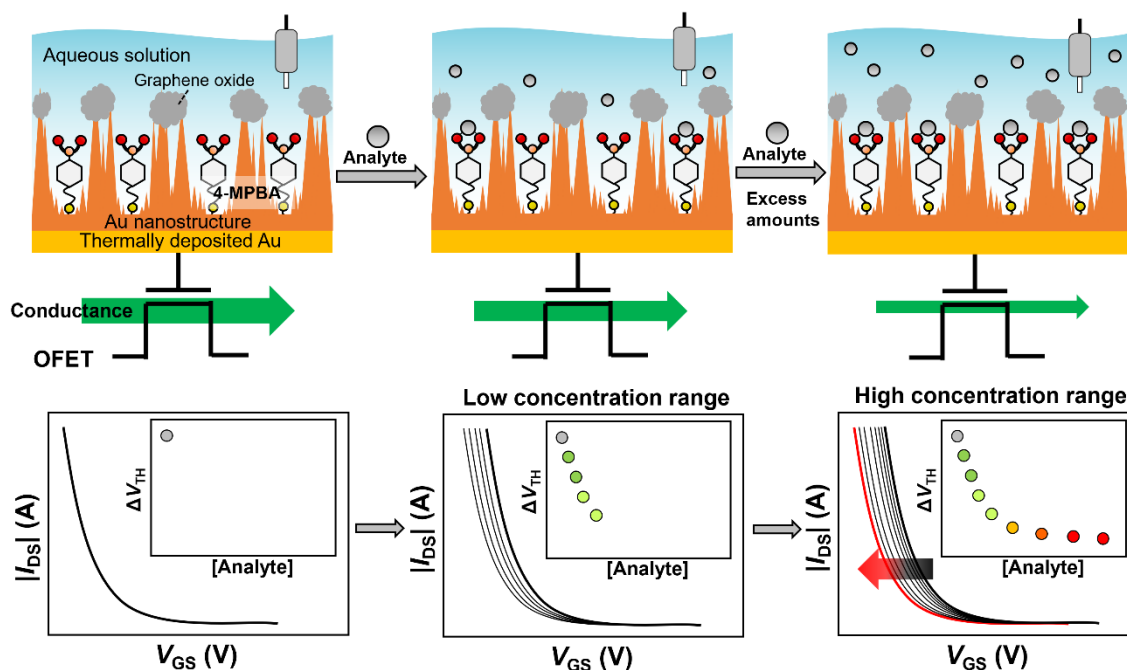
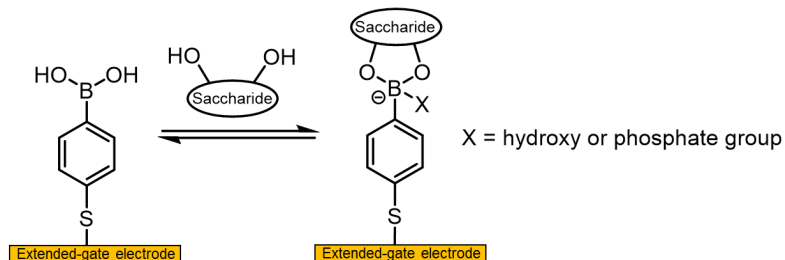


Fig. S6. Schematic illustration of the sensing mechanism of the extended-gate-type OFET-based sensor.

Saccharide detection mechanism of 4-MPBA

The boronic acid derivative (*i.e.*, 4-MPBA) on the extended-gate electrode reacts to cis-diol moieties of target saccharides, which form sp^3 -type boronate esters through dynamic covalent bonds (Scheme S1).⁵⁵ The negative boronate anions formed by the target saccharides cause the changes in the surface potential of the extended-gate electrode, which induce the shifts of transfer characteristics.



Scheme S1. Boronate esterification of 4-MPBA on the extended-gate electrode by adding saccharides.

Detection of glucose by the GO-OFET

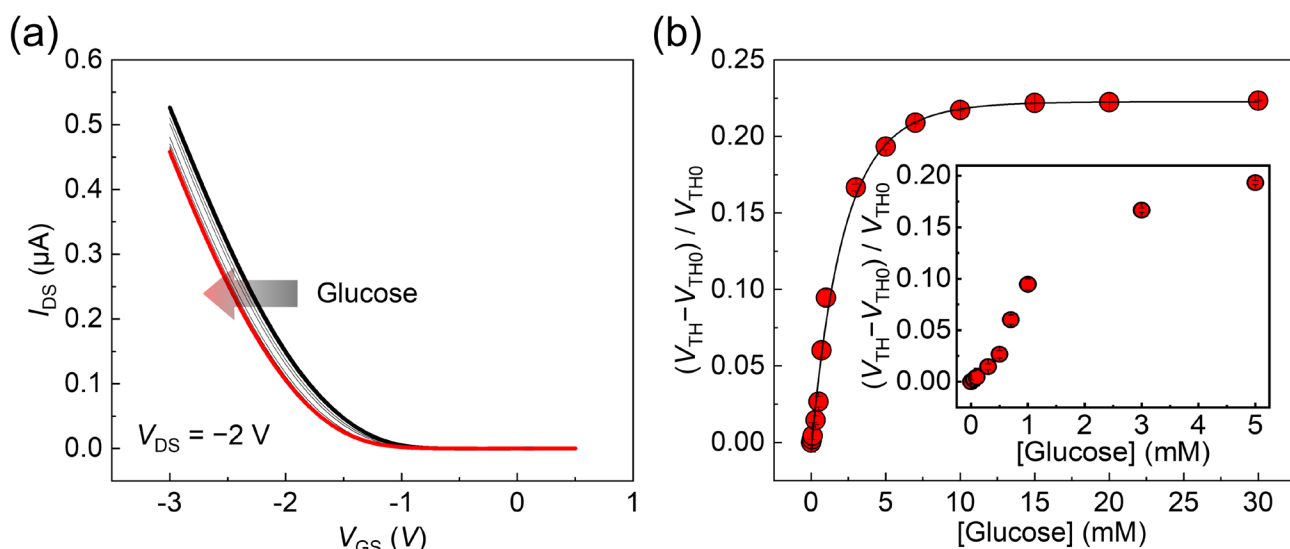


Fig. S7 (a) Transfer characteristics of the GO-OFET-based chemical sensor with 4-MPBA upon the addition of glucose in 0.1 M phosphate buffer with 0.1 M NaCl at pH 7.4. (b) The titration isotherm obtained by collecting V_{TH} at each concentration of glucose from 0 to 30 mM. The terms V_{TH0} and V_{TH} respectively indicate threshold voltages before and after adding glucose. Three repetitive evaluations were carried out for each concentration. The inset indicates the transistor response to glucose at the low concentration range from 0 to 5 mM.

Example of selectivity test by the GO-OFET

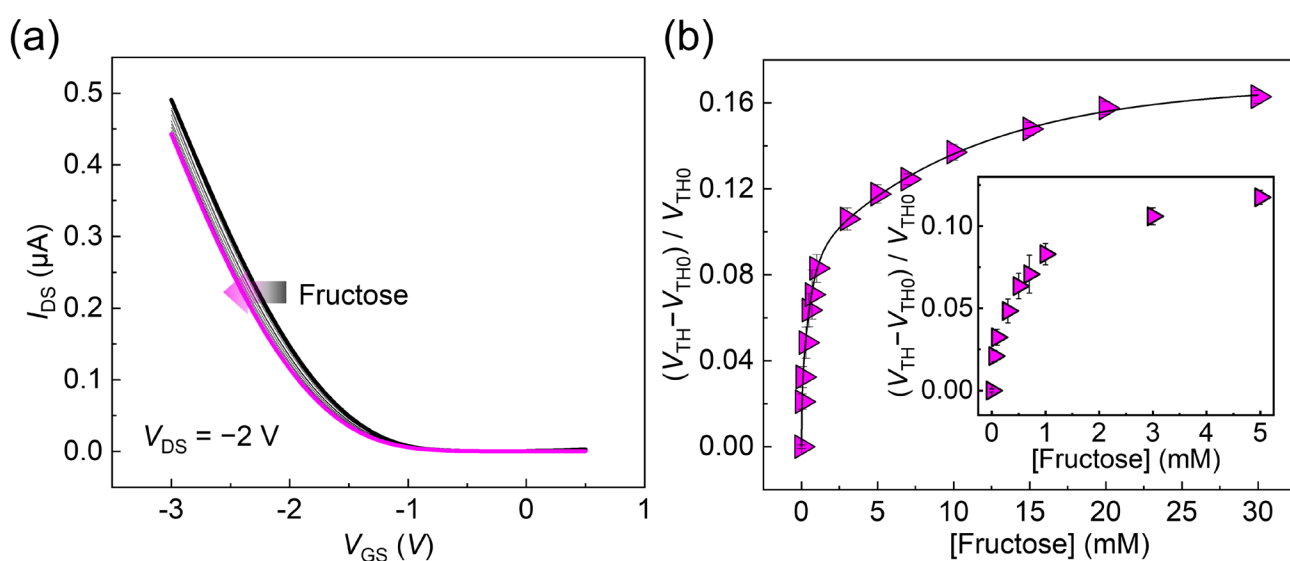


Fig. S8 (a) Transfer characteristics of the GO-OFET-based chemical sensor with 4-MPBA upon the addition of fructose in 0.1 M phosphate buffer with 0.1 M NaCl at pH 7.4. (b) The titration isotherm obtained by collecting V_{TH} at each concentration of fructose from 0 to 30 mM. The terms V_{TH0} and V_{TH} respectively indicate threshold voltages before and after adding fructose. Three repetitive evaluations were carried out for each concentration. The inset indicates the transistor response to fructose at the low concentration range from 0 to 5 mM.

Response to glucose by the OFET without GO

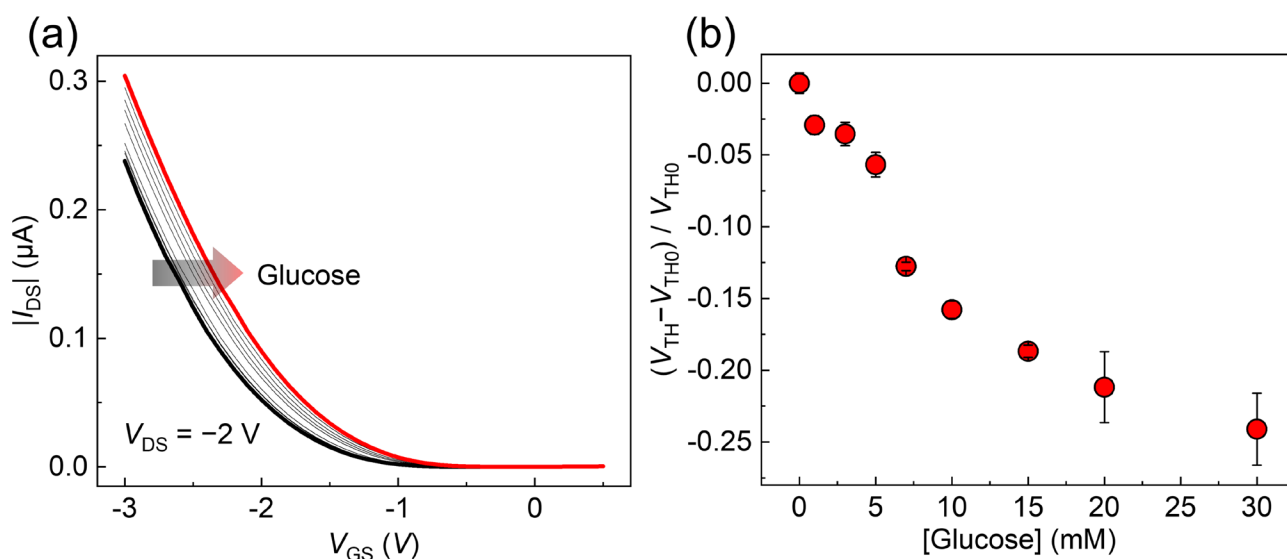


Fig. S9 (a) Transfer characteristics of the OFET-based chemical sensor with 4-MPBA upon the addition of glucose in 0.1 M phosphate buffer with 0.1 M NaCl at pH 7.4. (b) The titration isotherm obtained by collecting V_{TH} at each concentration of glucose from 0 to 30 mM. The terms V_{TH0} and V_{TH} respectively indicate threshold voltages before and after adding glucose. Three repetitive evaluations were carried out for each concentration.

Antifouling ability test for the extended-gate electrode

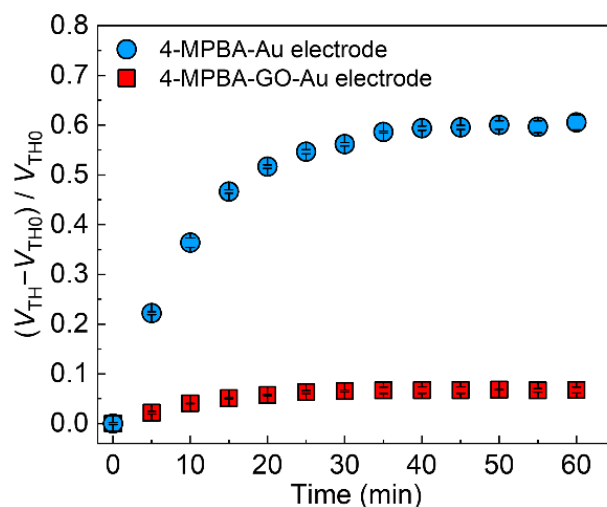


Fig. S10 Antifouling ability test for the OFET in 0.1 M phosphate buffer with 0.1 M NaCl (pH 7.4) containing negatively charged proteins (40 g/L serum albumin and 10 g/L immunoglobulin G). The red squares and blue circles mean the 4-MPBA-attached extended-gate Au electrode with and without GO, respectively. Three repetitive evaluations were carried out for each electrode.

5. Spike and recovery test for glucose in human blood plasma

Table S1 Estimated recovery rates of glucose in human blood plasma ($n=3$)

Spiked glucose (mM)	Found glucose concentration (mM)	Recovery rate (%)
0.0	4.3 ± 0.27	98
0.5	4.8 ± 0.57	87
1.0	5.5 ± 0.25	110
1.5	5.9 ± 0.83	102
2.0	6.3 ± 0.44	97
2.5	6.8 ± 0.58	94

References

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