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Supplemental information

**Peptide sequence-driven direct electron transfer
properties and binding behaviors of gold-binding
peptide-fused glucose dehydrogenase on electrode**

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Supplemental information

: Figure S1-S8 and Table S1 and S2

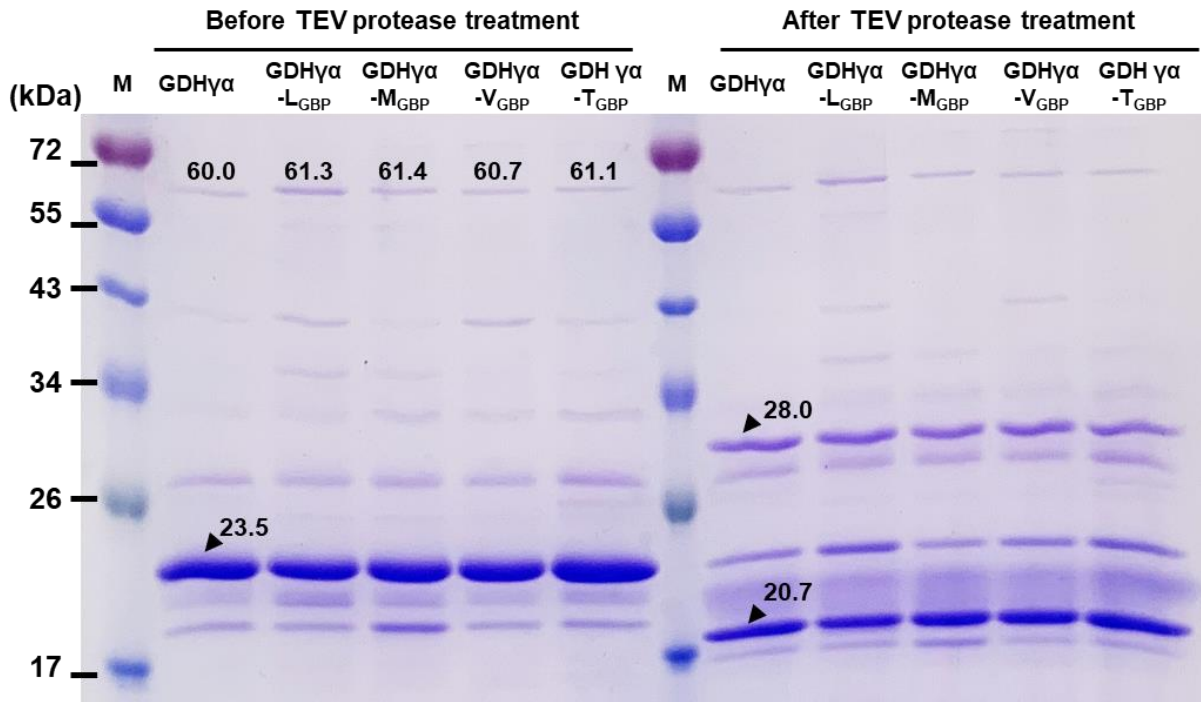
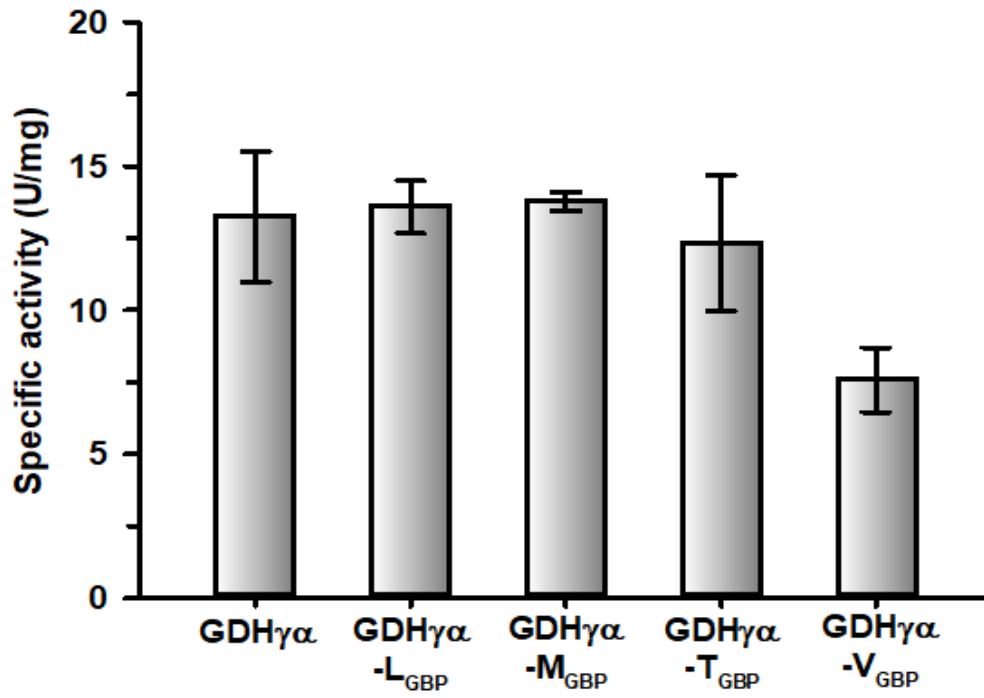


Figure S1. SDS-PAGE analysis of GDH α proteins (native GDH α and GDH α -X_{GBP}; X = L, M, T, and V) before and after TEV protease treatments. SDS-PAGE gel (12%); band near 60 kDa: GDH α or GBP-tagged GDH α ; band at 23.5 kDa: GDH γ before TEV protease treatment; band at 20.7 kDa: GDH γ after TEV protease treatment; band at 28 kDa: TEV protease. M: marker, related to Figure 1.

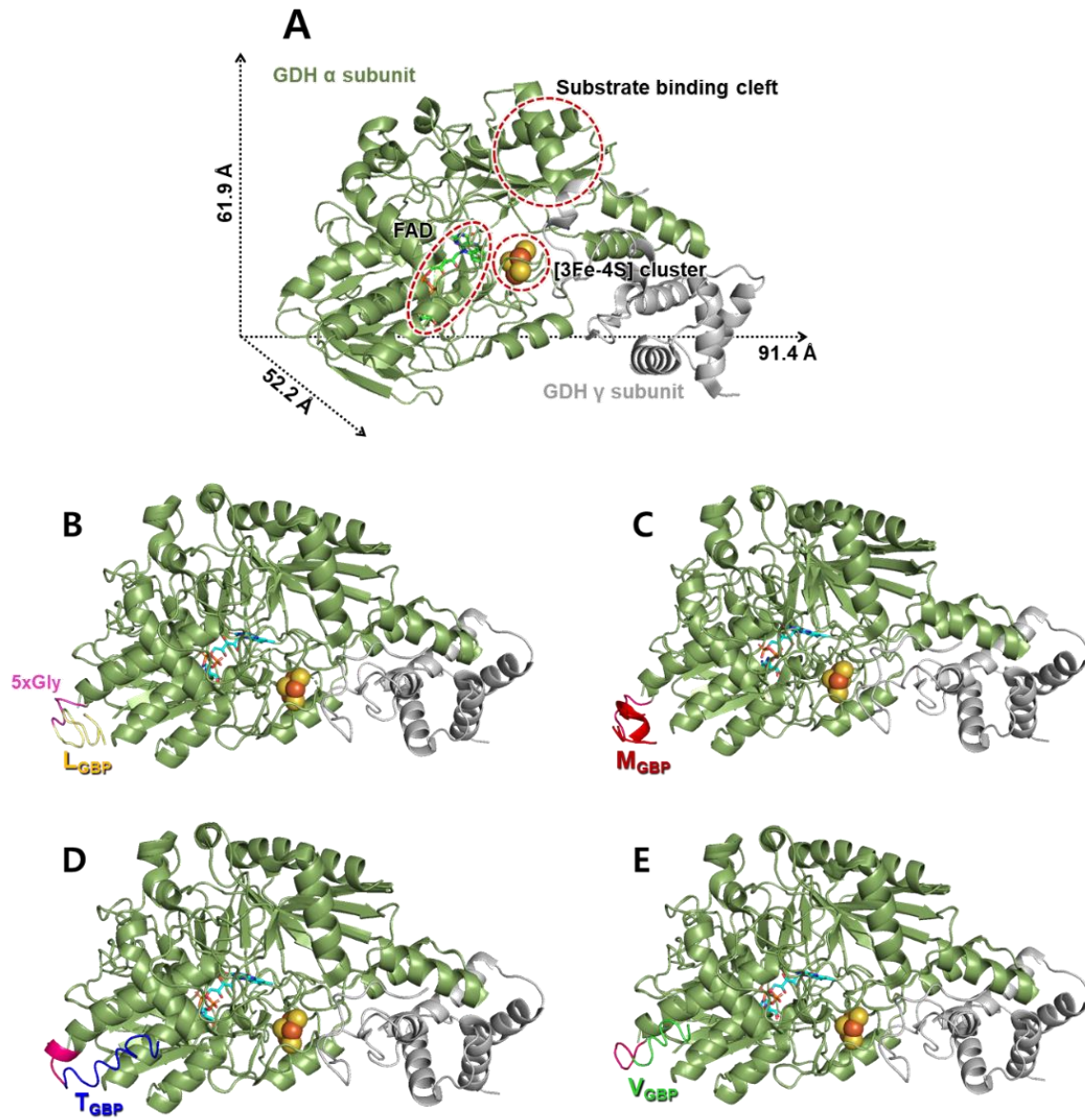


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11 **Figure S2. Comparison of enzyme activity of native GDH $\gamma\alpha$ and mutants (GDH $\gamma\alpha$ -X_{GBP}; X = L, M, T, and**
12 **V) according to glucose oxidation rate.** Reaction mixture consisted of 0.05 mg/mL protein, 100 mM glucose,

13 0.5 mM DCIP, and 6 mM PMS in 10 mM phosphate buffer (pH 7.4), related to Figure 1.

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16 **Figure S3. Prediction models of native GDH α and fusion GDH α proteins.** (A) Crystal structure of GDH α

17 complex reported previously (PDB ID: 6A2U). Herein, the protein structure is visualized with PyMOL program,

18 and the dimensions were analyzed in the same program. Structural models of (B) GDH α -LGBP, (C) GDH α -

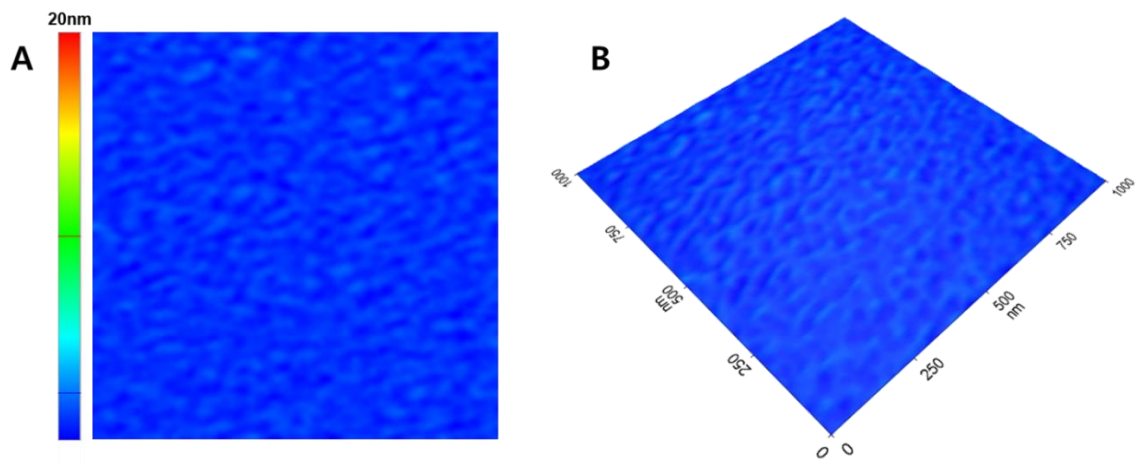
19 MGBP, (D) GDH α -TGBP, and (E) GDH α -VGBP. The structures of the GDH α subunit fused with different GBPs,

20 which presented highest C-value predicted by the I-TASSER, was aligned on PyMOL using previously

21 determined crystal structures for native FAD-dependent glucose dehydrogenase from *Burkholderia cepacia*

22 (PDB ID: 6A2U), related to Figure 2.

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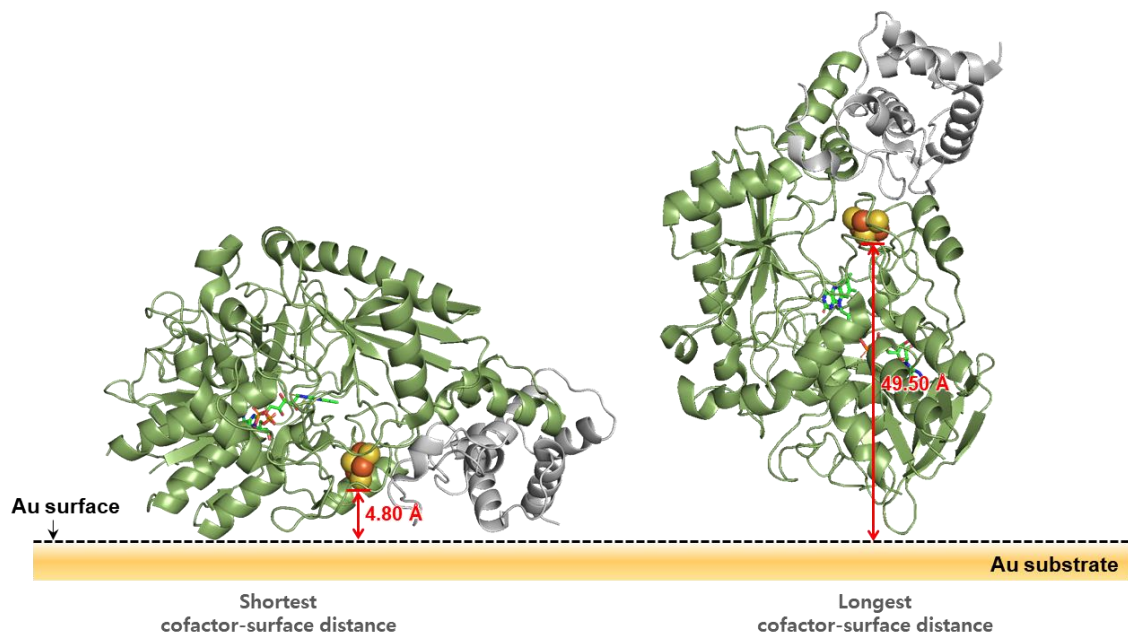


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25 **Figure S4. AFM images for bare Au surface used for enzyme binding studies. (A) 2-D topography and (B)**

26 **3-D topography; scanned area is $1\ \mu\text{m} \times 1\ \mu\text{m}$ related to Figure 4.**

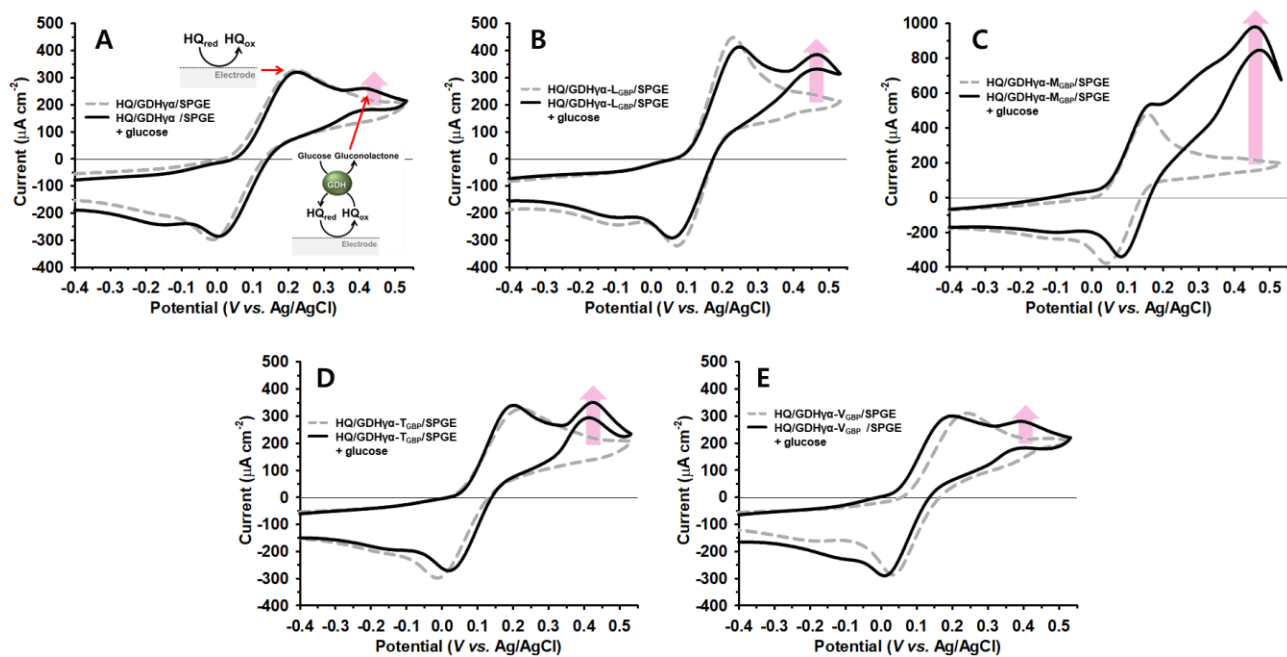
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29 **Figure S5. Schematic depiction of GDH α -bound gold substrate, and shortest and longest distance**
30 **between [3Fe-4S] cluster and electrode surface available according to enzyme binding orientation on**
31 **electrode, related to Figure 5.**

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34 **Figure S6. CV of mediated electron transfer systems.** The CV profiles of (A) HQ/GDH α /SPGE, (B)

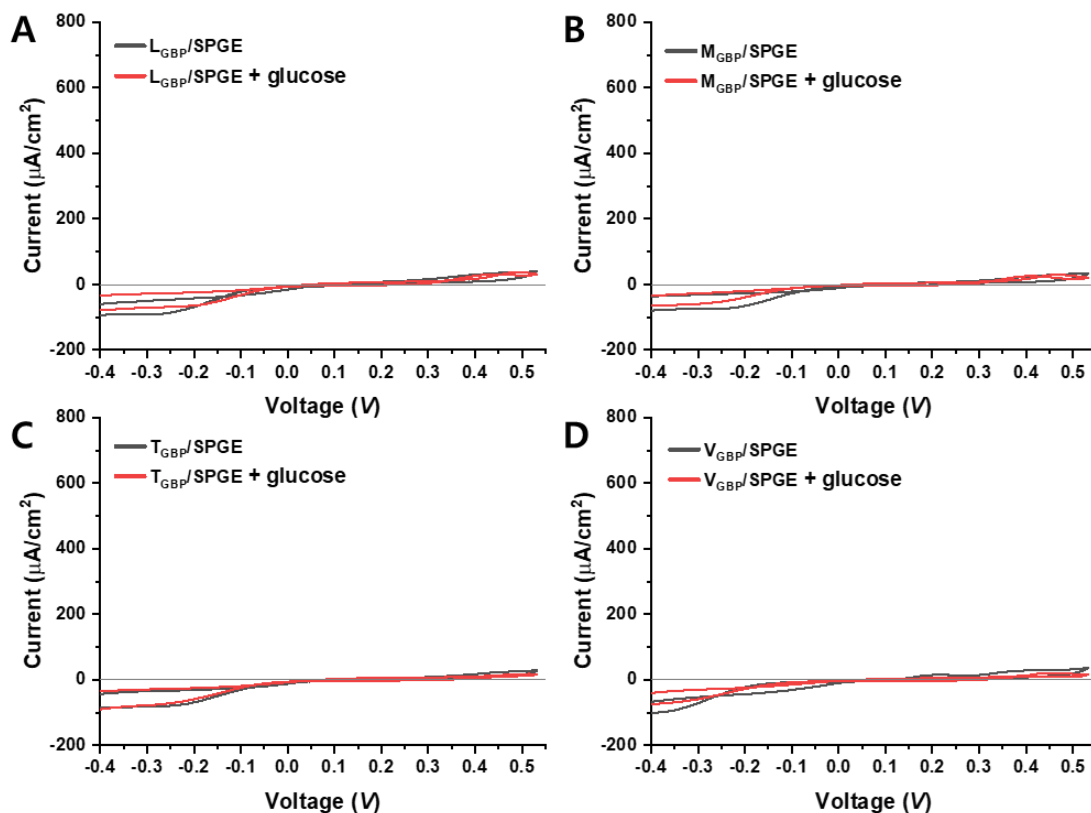
35 HQ/GDH α -L_{GBP}/SPGE, (C) HQ/GDH α -M_{GBP}/SPGE, (D) HQ/GDH α -T_{GBP}/SPGE, and (E) HQ/GDH α -

36 V_{GBP}/SPGE in the absence and presence of glucose 100 mM in PBS buffer (pH 7.4) (scan rate: 100 mV s⁻¹).

37 The inset within the (A) is schematic description of reaction mechanism corresponding with anodic peaks,

38 related to Figure 6.

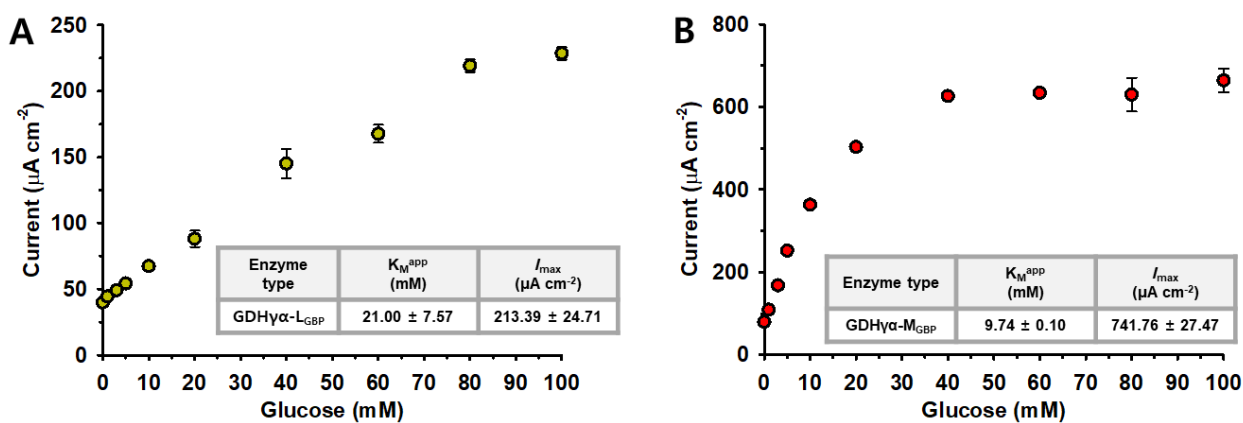
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41 **Figure S7. CV at the GBP-only conditions.** The CV profiles of (A) $L_{\text{GBP}}/\text{SPGE}$, (B) $M_{\text{GBP}}/\text{SPGE}$, (C) $T_{\text{GBP}}/\text{SPGE}$,
 42 (D) $V_{\text{GBP}}/\text{SPGE}$ in the absence and presence of glucose 100 mM in PBS buffer (pH 7.4) (scan rate: 100 mV s^{-1}). Each peptide was immobilized on electrode surface by immersing the SPGEs in the $50 \mu\text{M}$ peptide for 2hrs,
 43 ¹). Each peptide was immobilized on electrode surface by immersing the SPGEs in the $50 \mu\text{M}$ peptide for 2hrs,
 44 with mild shaking, related to Figure 6.

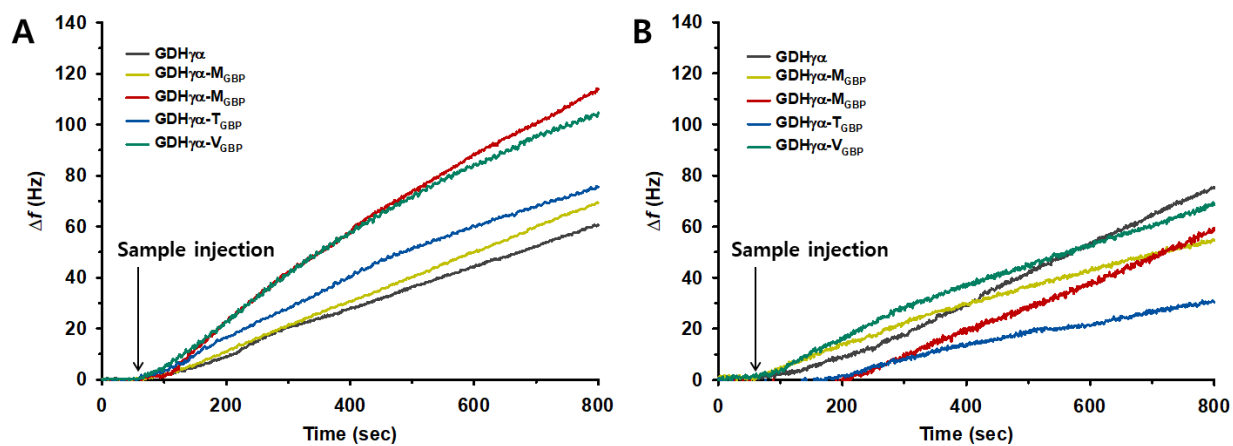
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47 **Figure S8. Plot of anodic peak current (I_{peak}) from cyclic voltammetry.** The I_{peak} of (A) GDH α -L_{GBP}/SPGE
 48 and (B) GDH α -M_{GBP}/SPGE was obtained during successive glucose injections (1, 3, 5, 7, 10, 20, 40, 60, 80,
 49 and 100 mM). Inset: apparent electrochemical parameters (K_M^{app} and I_{max}) of GDH α -L_{GBP}/SPGE or GDH α -
 50 M_{GBP}/SPGE, related to Figure 6.

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53 **Figure S9. QCM frequency changes after injection of wild type GDH $\gamma\alpha$ and GDH $\gamma\alpha$ -X_{GBP} (X = L, M, T, and**

54 **V) on (A) Au and (B) Si surfaces, related to Figure 9 and Figure 10.**

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56 **Table S1. Gold-binding peptides and peptide properties**, related to Figure 1.

GBP types	Sequence	Molecular weight (Da)	Isoelectric point	References
L_{GBP}	LKAHLPPSRLPS	1,315.58	11.41	Nam <i>et al.</i> , 2006
M_{GBP}	MHGKTQATSGTIQS	1,446.60	9.88	Brown, 1997
T_{GBP}	TGTSVLIATPGV	1,115.29	6.02	Huang <i>et al.</i> , 2005
V_{GBP}	VSGSSPDS	734.72	3.75	Kim <i>et al.</i> , 2010

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59 **Table S2. Onset and peak potentials in cyclic voltammogram of native GDH α /SPGE or GDH α -**
 60 **X_{GBP}/SPGE (X = L, M, T, or V), related to Figure 6.**

Types of enzyme-electrodes	E_{on}^a (V vs. Ag/AgCl)	E_p^b (V vs. Ag/AgCl)
GDHα/SPGE	0.364 \pm 0.006	0.420 \pm 0.006
GDHα-L_{GBP}/SPGE	0.036 \pm 0.009	0.359 \pm 0.011
GDHα-M_{GBP}/SPGE	-0.304 \pm 0.003	-0.091 \pm 0.005
GDHα-T_{GBP}/SPGE	N.M. ^c	N.M.
GDHα-V_{GBP}/SPGE	0.191 \pm 0.004	0.310 \pm 0.004

61 ^aonset potential; ^bpeak potential; ^cnot measurable