Supplementary Materials

Engineered Glucose Oxidase Capable of Quasi-Direct Electron Transfer after Quick-and-Easy Modification with a Mediator

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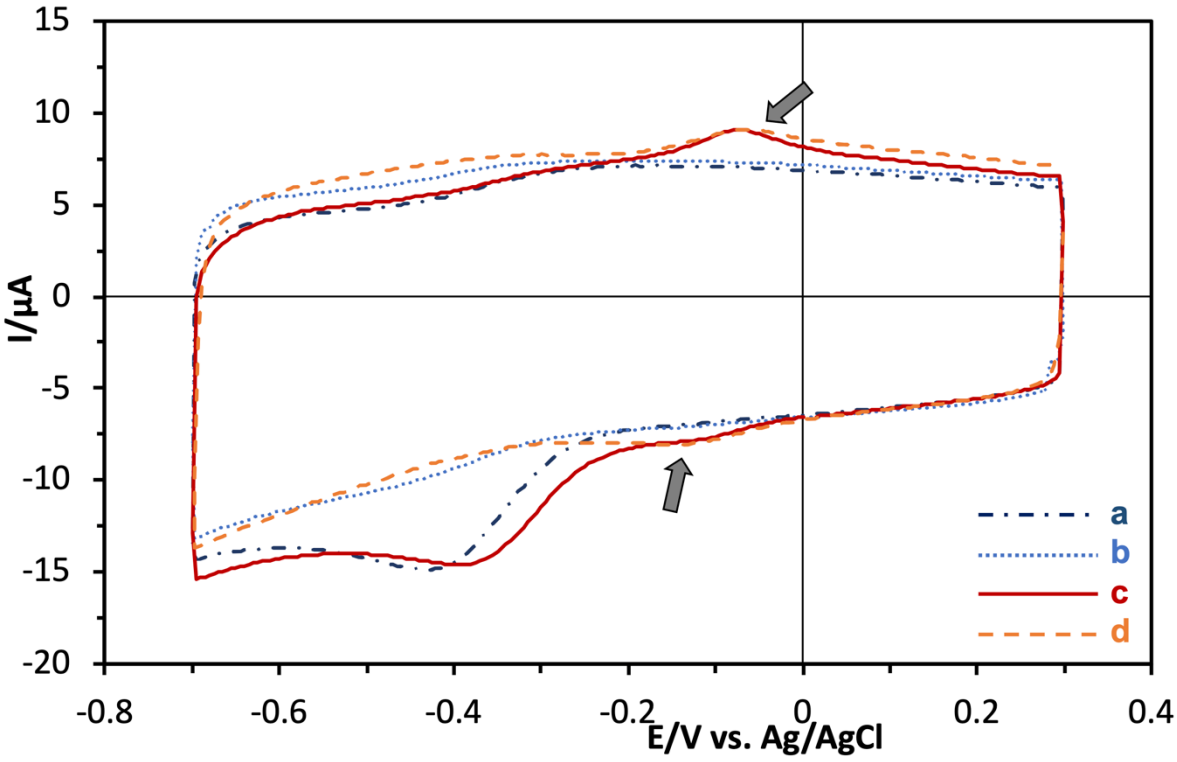

Figure S1. *Cyclic voltammograms of (a) unmodified AnGOx-WT in the absence of glucose; (b) unmodified AnGOx-WT in presence of 33 mM glucose; (c) PES-modified AnGOx-WT in absence of glucose; (d) PES-modified AnGOx-WT in presence of 33 mM glucose. Scan rate 50 mV/s. Arrows: peaks due to PES.*

Table S1. *Kinetic parameters of AnGOx-WT and AnGOx-I489K*

Enzyme	PES modification	Oxidase Activity			Dehydrogenase Activity		
		V_{max} (U/mg)	K_m (mM)	V_{max}/K_m (U/mg·mM)	V_{max} (U/mg)	K_m (mM)	V_{max}/K_m (U/mg·mM)
*An*GOx-WT	−	140	36	3.8	50	32	1.6
	+	170	40	4.3	53	26	2
*An*GOx-I489K	−	76	28	2.7	22	22	1
	+	130	42	3	49	23	2.2

Oxidase activity was determined by monitoring the increase of absorbance at 555 nm (formation of quinoeimine dye) of a mixture of GOx, glucose (var. concentrations), 1.5 mM 4-aminoantipyrine (FUJIFILM Wako Pure Chemical Corporation, Osaka, Japan), 1.5 mM *N*-ethyl-*N*-(2-hydroxy-3-sulfopropyl)-3-methylaniline (Dojindo Laboratories Co. Ltd., Kumamoto, Japan), and 2 U/mL horseradish peroxidase (HRP) (Amano enzyme Inc., Gifu, Japan) in 20 mM P.P.B.. Quinoeimine dye is formed by HRP in presence of hydrogen peroxide. The formation of 1 μmol/min hydrogen peroxide, corresponding to the oxidation of 1 μmol/min glucose, was defined as 1 U oxidase activity.

Dehydrogenase activity was determined by monitoring the decrease of absorbance at 600 nm (reduction of 2,6-dichlorophenolindophenol, DCIP (Kanto Chemical Co. Inc., Tokyo, Japan)) of a mixture of GOx, glucose (var. concentrations), 0.6 mM PMS, and 0.06 mM DCIP in 20 mM P.P.B. The reduction of 1 μmol/min DCIP, corresponding to the oxidation of 1 μmol/min glucose, was defined as 1 U dehydrogenase activity.