A Plant-Insertable Multi-Enzyme Biosensor for the Real-Time Monitoring of Stomatal Sucrose Uptake

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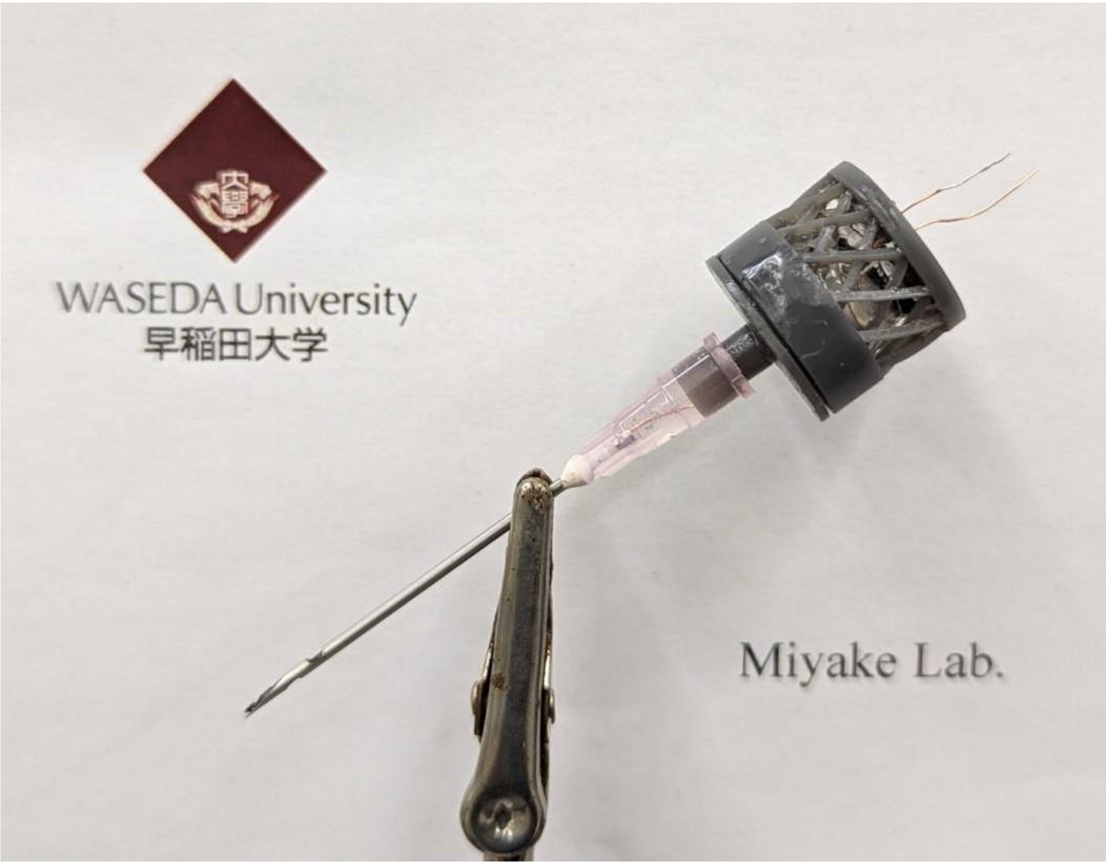

Figure S1. Photograph of needle type sucrose sensor prepared in this work.

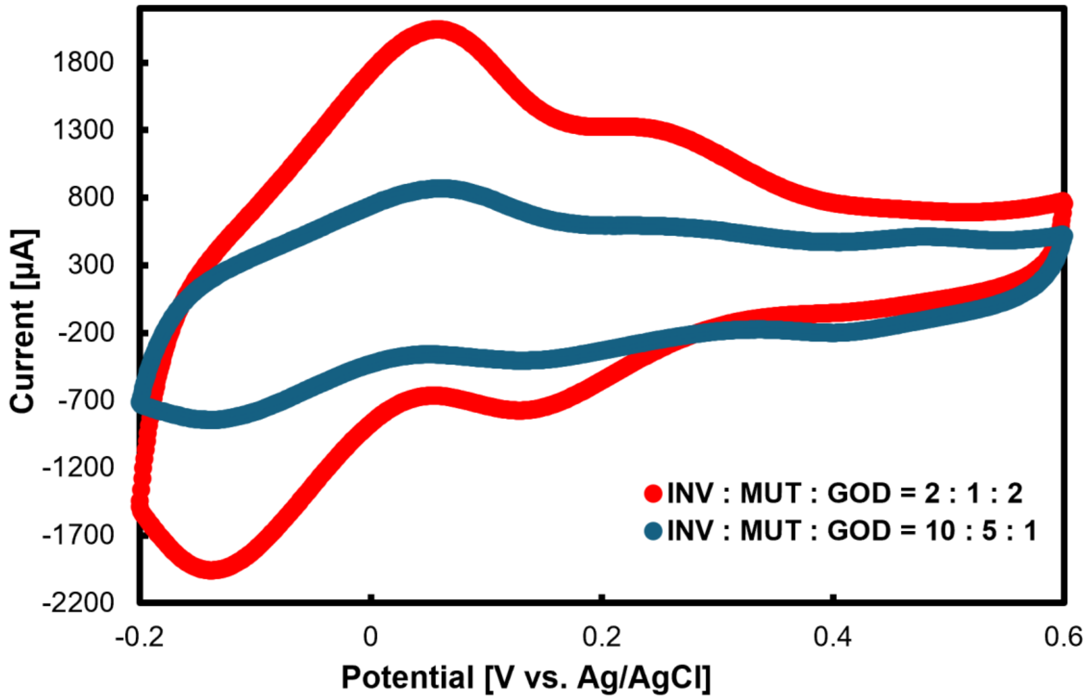

Figure S2. Cyclic voltammetry curves of anode with enzyme ratio INV : MUT : GOD = 2 : 1 : 2 (red) and 10 : 5 : 1 (blue) in 15 mM sucrose-PBS solution.

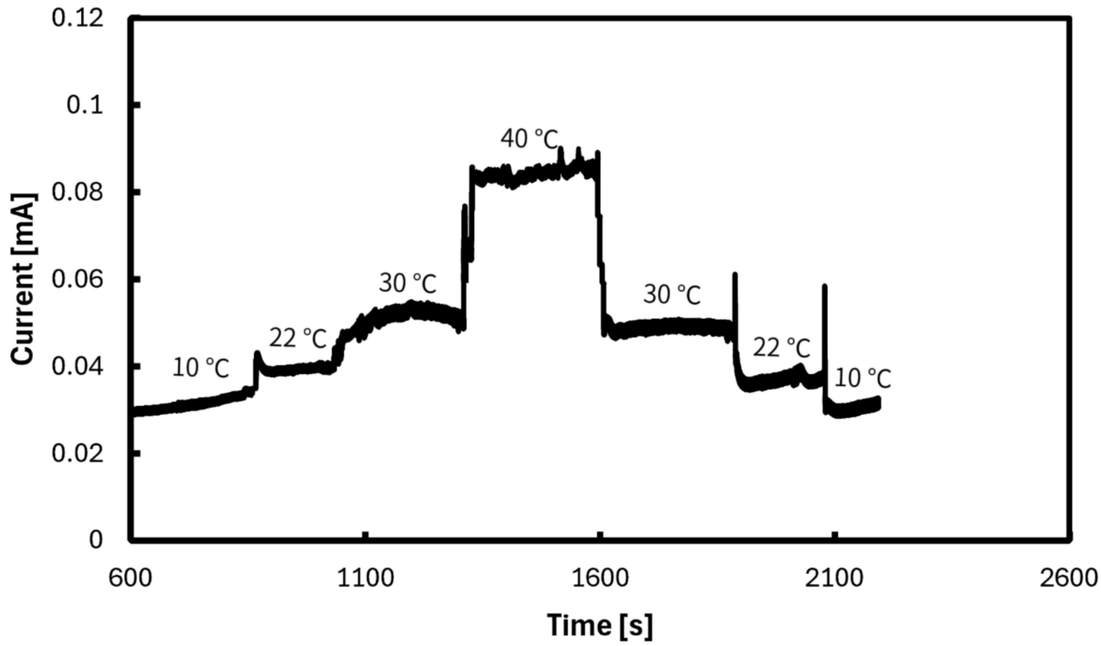

Figure S3. Stability evaluation of sensor response under repeated temperature cycling (10 °C → 22 °C → 30 °C → 40 °C → 30 °C → 22 °C → 10 °C).

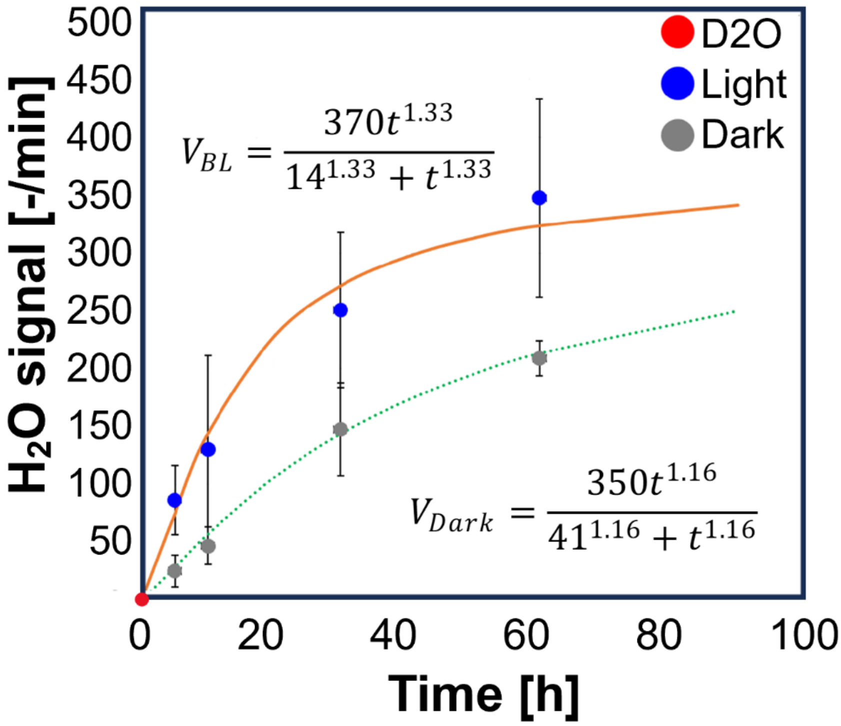

Figure S4. Efflux of H_2O molecules from intact Sugi leaves immersed in D_2O under light and dark conditions. Data were fitted using a Michaelis-Menten-like Hill-type equation. Each point is the average, and the error bars are the standard deviation between three samples (n = 3).

Figure S5. Photograph of needle type sucrose sensor inserted into Japanese cedar.

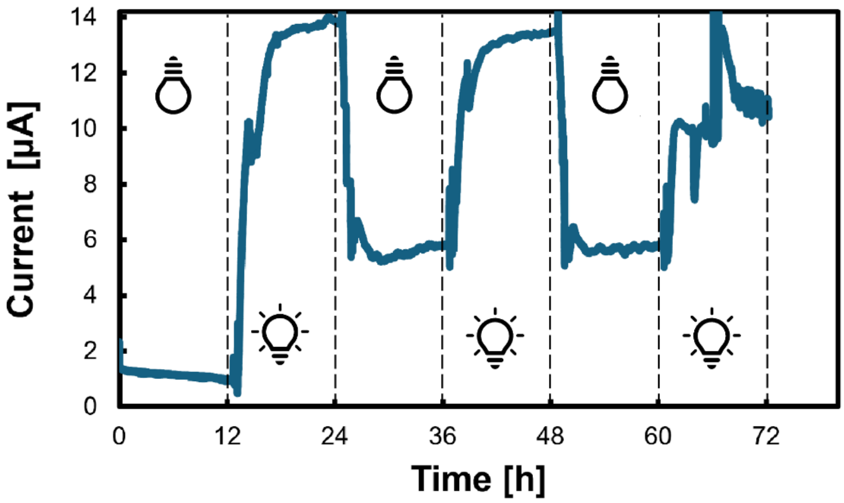

Figure S6. 72 hours real-time chronoamperometric (V = +0.6V vs BOD biocathode) monitoring of sucrose concentration in Japanese cedar soaked in 50 mM sucrose under different light conditions (0–12 h: light-off, 12–24 h: light-on, 24–35 h: light-off, 36–48 h: light-on, 48–60 h: light-off, 60–72 h: light-on).

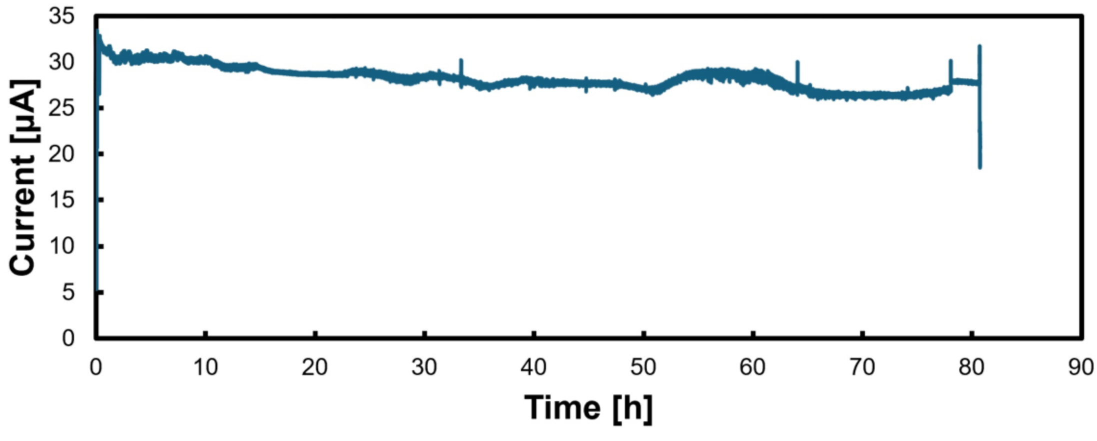

Figure S7. Operational stability of the sensor evaluated in 25 mM sucrose solution over 72 hours.

Table S1. Wide use of sensors on plants among published works and our work.

	Chen et al., 2024	Dufil et al., 2024	Diacci et al., 2020	Diacci et al., 2021	Taniguchi et al., 2020	Perdomo et al., 2023	**This work**
Sensitivity	17 nA μM^{-1} cm^{-2}	NA	NA	NA	NA	22.7 nA μM^{-1} cm^{-2}	**6.22 μA mM^{-1} cm^{-2}**
Response time	NA	NA	< 1 min	< 10 min	NA	< 10 min	**< 90 s**
Lifetime	> 12 h	< 1 day (GDH-FAD) > 2 day (GOx)	60 min	Several days	1500 h	Not available (NA)	**> 72h**
LOD	33.3 μM	100 μM (GDH-FAD) 1 mM (GOx)	NA	NA	μV level	9.7 μM	**100 μM**
Measured parameter	Glucose	Glucose	Glucose	Sucrose, glucose	Biopotential	Glucose	**Sucrose**
In-vivo	Yes	Yes	No	Yes	Yes	Yes	**Yes**
Real-time	Yes	Yes	No	Yes	Yes	No	**Yes**
Sensor placement	Microneedle patch on leaf and stem	Biohybrid root electrode	OECT probe in chloroplasts solution	OECT probe in stem cut	Sheet on leaf	Magnetic patch on leaves	**Needle on branch/stem**

References

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