Supporting Information

Flavin-Sensitized Electrode System for Oxygen Evolution Using Photo-Electrocatalysis

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Flavins

Lumiflavin (LF) was purchased from Sigma-Aldrich and flavin adenine dinucleotide (FAD) was from Boehringer Mannheim GmbH- Germany.

Voltammetric Measurements

Voltammetric measurements were undertaken in a conventional three-electrode cell at room temperature using a CHI 760 potentiostat. Ag|AgCl and high surface area Pt wire were used as reference and auxiliary electrodes, respectively. Prior to undertaking experiments, 0.5 M K_2HPO_4 / KH_2PO_4 aqueous buffer as the supporting electrolyte solution (pH 7.1) was de-aerated by purging with N_2 for at least 20 min. During measurements, a N_2 atmosphere was maintained inside the cell. All measurements were performed at room temperature. When applicable, illumination was provided by either a halogen desk lamp or a blue LED (intensities ~ 25mW/cm^2 at the sample). The experimental cell was kept at a distance from the light source to prevent any sample heating.

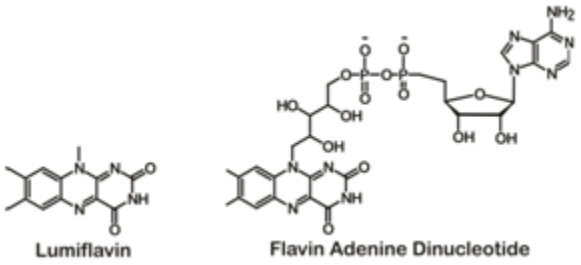
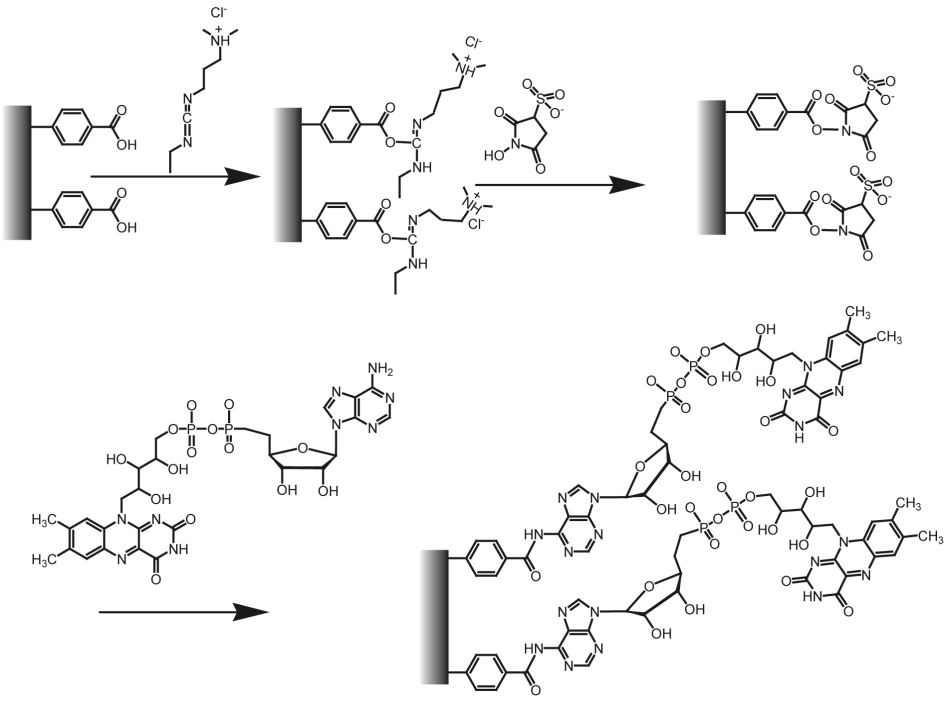


Figure S1: Structures of the model flavins: lumiflavin and FAD

Covalent grafting of FAD

Pencil lead electrodes were obtained from Graphtek LLC (Graphite Store), IL, USA (AE001535 and NC001290). Modification of the graphite electrodes (**GE**) with carboxyphenyl group was achieved by one-electron reductive adsorption of the carboxyphenyl diazonium salt in

0.1M HCl solution at room temperature.[1] Specifically, the electrodes were treated with 5 mM 4-carboxy phenyl diazonium tetrafluoroborate in 0.1M HCl for 12 hours. Formation of N_2 bubbles on the surface of the electrode is a positive sign of the diazonium grafting. In the second step, the carboxyl-functionalized electrodes were coupled to the FAD via a carbodiimide coupling reaction following a reported protocol.[2] Specifically, the electrodes were soaked in 5 mL of 0.1M MES buffer (pH 5.9) containing 0.2% Triton X-100, 40 mM EDC (N'-(ethylcarbonimidoyl)-N,N-dimethylpropane-1,3-diamine) and 40 mM sulfo NHS (*N*-hydroxysulfosuccinimide) for two hours to allow activation of the carboxylate groups. After two hours the electrodes were rinsed with a phosphate buffer solution (pH 7.2) and then treated with 1mM FAD solution in phosphate buffer solution (pH 7.2) for 12 hrs. Finally, the electrodes were washed with KPi (inorganic phosphate buffer, 0.5M, pH 7.1) buffer and then deionized water to remove any unbound FAD. A schematic is provided as Scheme S1.



Scheme S1: Steps involved in covalent grafting of **FAD** to **GE**.

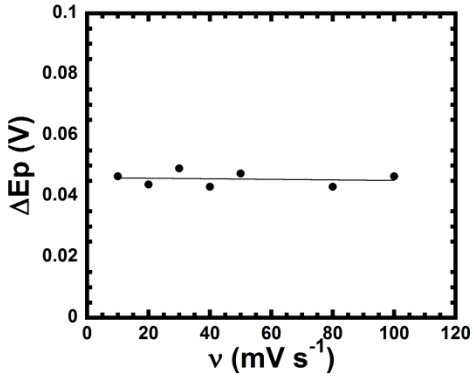


Figure S2: ΔE_p *vs*. scan rate for **LF**-functionalized **GE**.

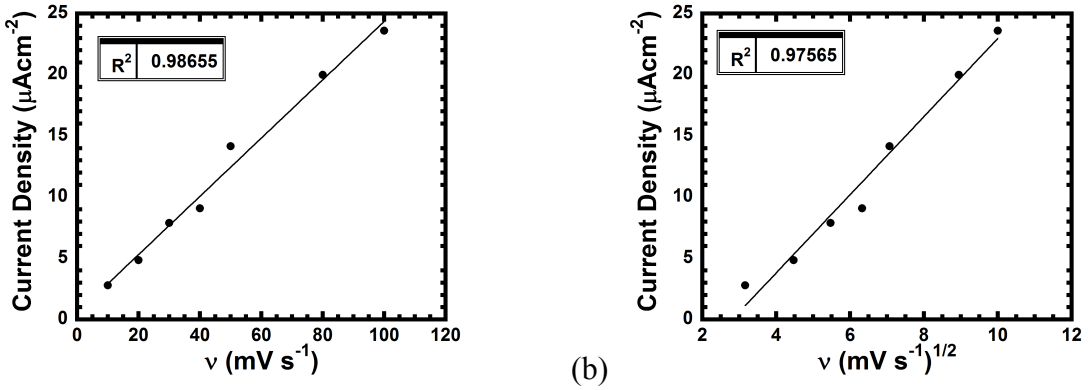


Figure S3: (a) Dependence of peak current at -0.48V on CV scan rates, (b) dependence of peak current on square root of CV scan rate. The plot of current vs. scan rate (a) shows better linearity than that of current vs. square root of scan rate (b), indicating successful immobilization of **LF** on the **GE** surface.[3]

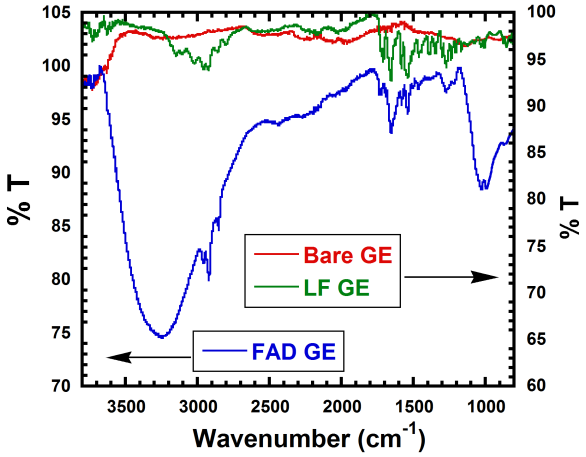


Figure S4: ATR-FTIR of bare **GE** (red line), **FAD**-functionalized **GE** (blue line) and **LF**-functionalized **GE** (green line). The flavin C=O stretches are near 1676 cm^{-1} and 1708 cm^{-1} respectively.[4] The C=N mode occurs between 1534 and 1548 cm^{-1} for bent through flat flavin.[5] Comparable peaks were observed for both **LF-GE** and **FAD-GE** but were absent for the bare **GE.** Note that the peaks assigned to N-H stretching are expected near 3300 cm^{-1} for free **FAD**. They are absent for the **FAD-GE**, as the amine group was converted to an amide as part of the covalent coupling to **GE**. The large broad feature extending from 2500-3600 cm^{-1} is due to water in the **FAD-GE** sample.

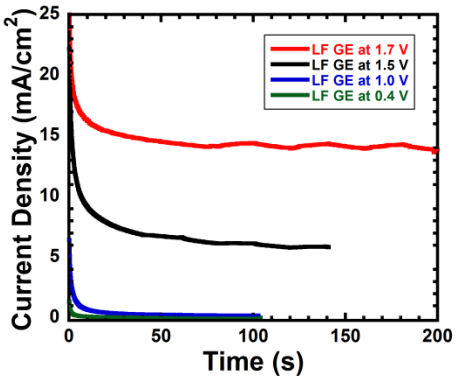


Figure S5: The i-t amperometric response to illumination for a **LF-GE** at different electrode potentials in 0.5M KPi buffer at pH 7.1. The ripples in the lines at 1.5 V and 1.7 V are the results of turning the visible light on (causing upward slopes) and off (causing downward slopes).

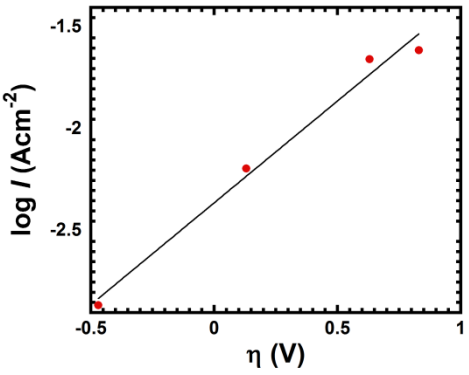


Figure S6: Tafel plots of current density as a function of overpotential for a **LF-GE**.

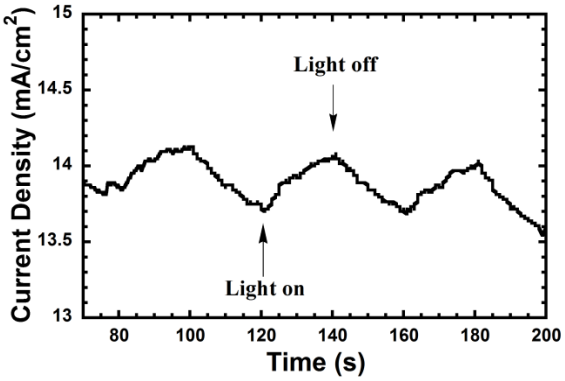


Figure S7: The i-t amperometric response of a **LF-GE** at a potential of 1.7 V in the presence or absence of light at pH 7.1.

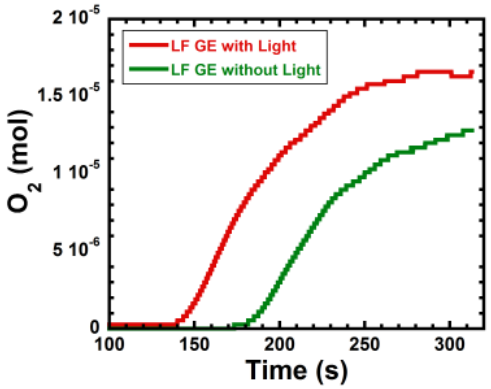


Figure S8. O_2 evolution curves recorded using a Clark-type O_2-sensing electrode. Conditions: Applied potential, 1.8 V; 4 mL 0.5 M KPi buffer, pH7.1 and room temperature. All conditions were the same for light and dark experiments, with the exception of whether or not illumination was applied.

In both the cases the electrolysis was started at 100 s however the O_2-sensing electrode was approximately 1 cm from the **LF-GE** where O_2 evolution was occurring, so a delay is expected for diffusion of O_2 from the site of formation to the site of detection. We also note that because formation of O_2 from H_2O is a multi-electron reaction, part of the lag time may also represent formation and accumulation of at least one intermediate prior to O_2 evolution. Similar lag times were previously observed during catalysis by inorganic complexes.[6] The lag time was shorter under illumination, suggesting that formation of at least one intermediate is accelerated by light in the presence of flavin (red line). However the onset voltage was not different in darkness vs. light arguing for the existence of at least one light-insensitive step as well. Ongoing efforts seek to clarify the identities of the steps and intermediates involved, and will be described in a later more lengthy report. However our CV measurements were conducted after the lag phase so the higher current at 1.4 V under illumination characterizes the illuminated state and corroborates our inference of faster formation of an oxidized species that leads to O_2, under the visible light. Indeed, the light-induced change in current (Figure S7) occurred on the same time-scale as the difference between the two lags.

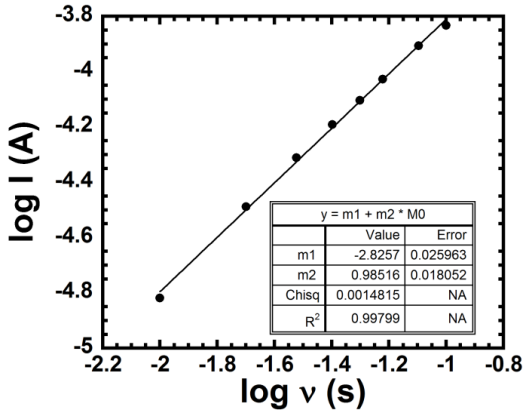


Figure S9: Plot of log I_{pa} vs. log ν for cyclic voltammograms of a **FAD-GC** electrode generated by covalent coupling. The slope of 0.98 suggests successful attachment of **FAD** to the surface. Scans were conducted in 0.5M KPi buffer at pH 7.1 and room temperature.

To assess the durability of the **LF-GE** electrodes, cyclic voltammetric scans were performed up to an anodic potential of 2.0 V. The electrodes were found to be significantly active up to more than ten consecutive cyclic scans. These electrodes were also active after storage under normal laboratory conditions for approximately two months (in darkness). As already mentioned in the body of the manuscript, the **LF-GE** electrodes showed considerable tolerance to highly oxidative potentials. Figure S10 depicts the performance of a **LF-GE** electrode after ten consecutive scans to +2.0 V with the persistence of the **LF** signal (as indicated by the cathodic peaks at -0.5V). However, the electrodes deteriorated gradually after extensive scanning to high anodic potential (20-25 scans to 2.0 V). Figures S11 **a** and **b** show the redox signal centered around $E^{\circ\prime} = -0.48$ V (vs. Ag/AgCl in 1M KCl) that reports on immobilized **LF** after exposure to high anodic potential, for two independent electrodes. After the first few (2-3) cycles to 2.0 V there was little difference in the cathodic current corresponding to **LF** (based on cathodic current at ~ -0.52 V). However ~40% of the immobilized **LF** was lost or modified after twenty anodic cycles up to 2.0 V (based on the cathodic current at ~ -0.5 V, Figure S12a). FT-IR spectra of the **LF-GE** electrodes recorded before and after using them for electrocatalytic water oxidation for 5-7 cycles are identical in the features they display, arguing against chemical modification of immobilized LF. However the IR signal intensities attributable to **LF** (1600 – 1200 cm^{-1}) were significantly attenuated after ~ 20 consecutive scans (Figure S12b). Thus we interpret the gradual loss of performance to detachment of **LF** from the electrodes for example as a result of oxidation to species that are more water-soluble than the initially-deposited **LF.**

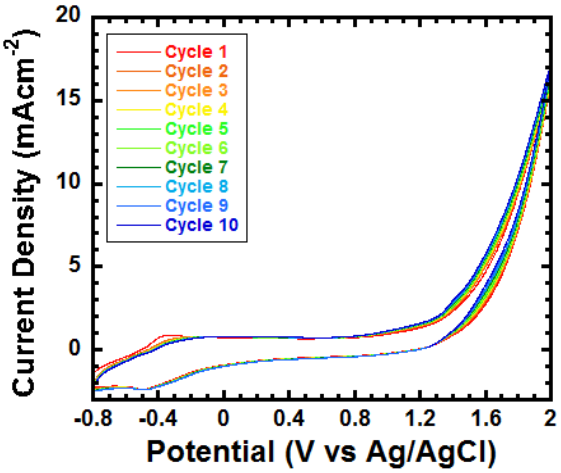
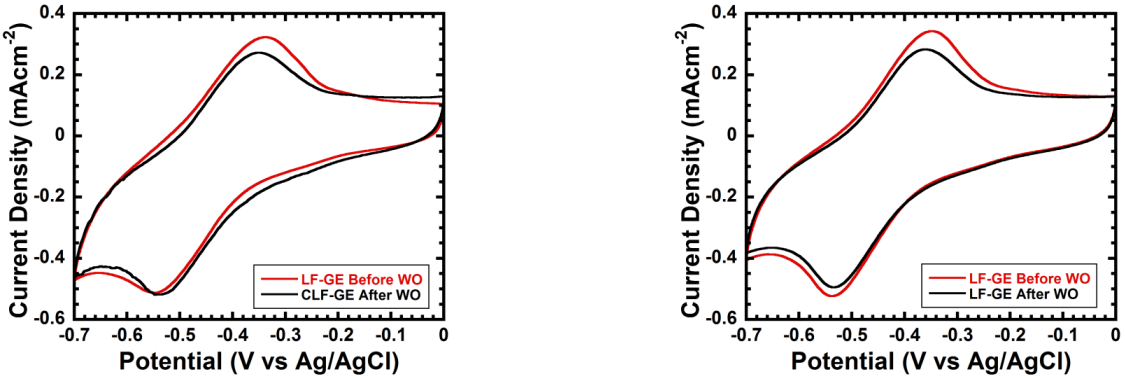
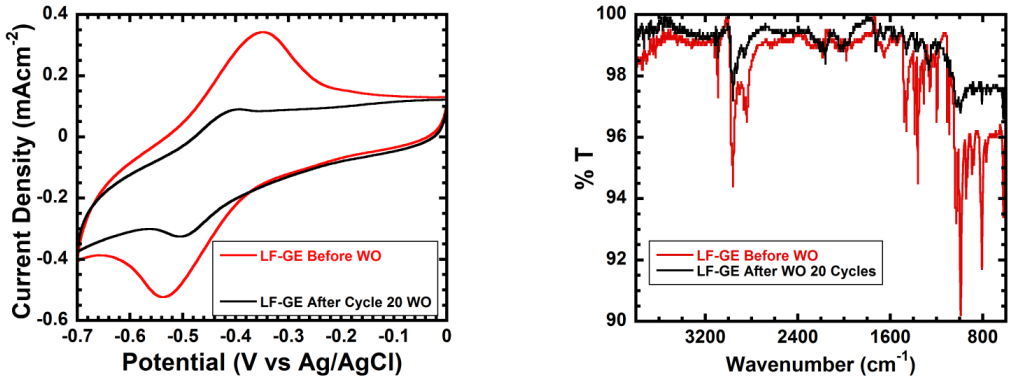


Figure S10: Cyclic voltammograms of **LF-GE** electrodes (two independent electrodes for figures **a** (two months old) and **b** (freshly fabricated)) showing the effect of repeated scanning. Conditions: scan rate, 50 mV/s; 0.5 M KPi buffer, pH 7 at room temperature. Electrodes are thus found to be stable at normal laboratory conditions for months when stored in darkness.



(a) **(b)**

Figure S11: Cyclic voltammograms of **LF-GE** electrodes (two independent electrodes for figures **a** and **b**) showing the presence of immobilized **LF** on the electrode before and after scanning up to high potentials (up to 2.0 V, two complete cycles, followed by purging the solution with N_2 to remove O_2 before recording 'after' CV). Conditions: scan rate, 20 mV/s; 0.5 M KPi buffer, pH 7 at room temperature.



(a) **(b)**

Figure S12: Cyclic voltammograms **(a)** and FT-IR **(b)** of **LF-GE** electrodes showing accrued deterioration of the electrodes after repetitive scanning up to 20 cycles to high potentials (up to 2.0 V). Electrochemical Conditions: scan rate, 20 mV/s; 0.5 M KPi buffer, pH 7 at room temperature. 'After WO' electrodes had to be oven-dried for 40 min at ≈ 50 °C in preparation for collection of IR data, which also has potential to affect the integrity of the LF and the electrode.

We also assessed the stability of the **LF-GE** electrodes towards oxidation of water at high anodic potential up to 2.0 V under illumination. Once again, the electrodes were found to retain substantial activity towards water oxidation and remain mechanically sound over multiple cycles. Figure S13 shows stability of a **LF-GE** electrode (~ 2 months old) after 14 consecutive scans with minimal change of the **LF** signal (cathodic peak at -0.5V). Interestingly, the anodic peak at 1.4V grew in successive scans under illumination to become *more* prominent. These data further supports our hypothesis that a transient flavin radical cation, capable of water oxidation, is formed under the reaction conditions.

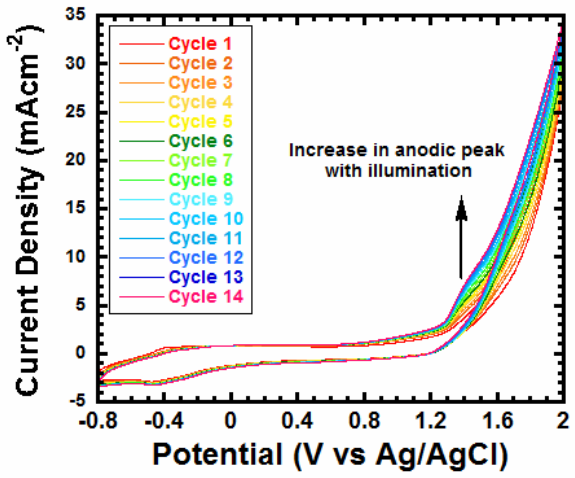
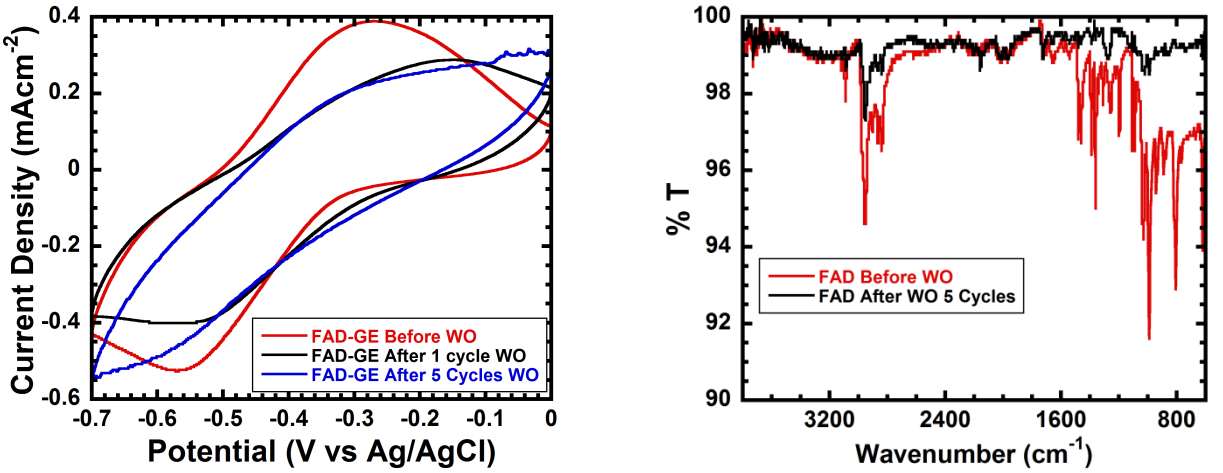


Figure S13: Cyclic voltammograms of **LF-GE** electrodes (~2 months old) showing the effect of repeated scans under illumination. Conditions: scan rate, 50 mV/s; 0.5 M KPi buffer, pH 7 at room temperature. Formation of the reactive species (proposed to be a transient flavin-derived radical cation) is prominent at ~1.4V upon repeated scanning.

As noted in the body of the manuscript, the **FAD**-functionalized graphite electrodes suffered from unsatisfactory durability, becoming inactive after only 5 cycles. CV scans in the range of -0.8V to 0.0 V were run before and after using **FAD-GE** for water oxidation (-0.8V to 2.0 V) in order to assess retention of FAD. The redox signal centered near $E^{\circ\prime} = -0.48$ V (vs. Ag/AgCl in 1M KCl) was measured as a signature of **FAD** immobilized on the electrode surface. As shown in Figure S14a ~25% of the immobilized **FAD** (based on the cathodic current at ~-0.56 V) was modified or lost after the first cycle up to 2.0 V and back. After five such cycles the electrode had significantly deteriorated as indicated by the almost-complete disappearance of the **FAD** signal (accompanied by the change in the capacitive current of the electrode, Figure S14a). The electrodes were further characterized by FT-IR spectroscopy before and after using them for electrocatalytic water oxidation. As seen from the comparison of the FT-IR spectra (Figure S14b) the signal intensities attributable to **FAD** (1600 – 1200 cm^{-1}) were either significantly diminished or missing. We hypothesize that flavin loss from these electrodes can be attributed to the relatively high water solubility of FAD in combination with possible hydrolysis of the phosphate linker that ties the flavin to the adenine which is coupled to the graphite. Our ongoing

efforts in this area seek to develop conductive polymer matrices to achieve improved entrapment of flavin molecules and provide enhanced electrical contact with the electrode.



(a) **(b)**

Figure S14: Cyclic voltammograms **(a)** and FT-IR **(b)** of **FAD-GE** electrodes showing deterioration of the electrode after repeated scans to high potentials (up to 2.0 V). Electrochemical Conditions: scan rate, 20 mV/s; 0.5 M KPi buffer, pH 7 at room temperature.

References

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