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HYBRID ENZYME-BIMETALLIC NANOPARTICLE SYSTEM FOR TANDEM OXIDATION CATALYSIS

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Based on previous work, I worked to develop a synthetic method for synthesizing bimetallic nanoparticles using glucose oxidase (GOx), where GOx is bound to the surface of the metallic nanoparticles. Previous work in literature and the research group indicates that adding the metallic precursor and GOx without the need for another reducing agent produces monometallic Au nanoparticles (AuNP) and gold-copper nanoparticles (AuCuNPs). The GOx acts as both the reducing and stabilizing agent. A goal of this project is to use the catalytic potential of the metallic nanoparticles and the enzymes to perform reactions in tandem. Prior work in the Nigra research group has successfully paired AuNPs and GOx to perform the sequential oxidation of glucose to gluconic acid (mono-acid) and then to glucaric acid (di-acid) products. This work has two primary aims. The first aim is to develop a new method of directly synthesizing bimetallic Au alloy nanoparticles using enzymes as both reducing and stabilizing agents. The second aim is to determine the effect of different synthetic procedures on the structure and function of the enzyme-metal nanoparticle hybrid material.

Using GOx to reduce and stabilize metal NPs, I investigated the catalytic activity of this material. Prior work in the group has demonstrated when the enzyme is added to the already reduced AuNPs by sodium borohydride, that the activity of the GOx is preserved. During the initial investigation of the catalytic systems, I measured the activity of the enzyme. The enzymatic reaction, where GOx consumes glucose to produce gluconic acid and hydrogen peroxide, can be approximated by measuring the rate of production of hydrogen peroxide. This was done though an enzymatically catalyzed reaction, where 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid), or ABTS, reacts with hydrogen peroxide using horseradish peroxidase (HRP) to produce a green product. The rate of production for the green ABTS acts as a proxy for the rate of production of hydrogen peroxide, and therefore the rate of the GOx reaction. The formation of this ABTS product is easily recorded using a UV-Vis spectrophotometer and measuring absorbance at 420nm.

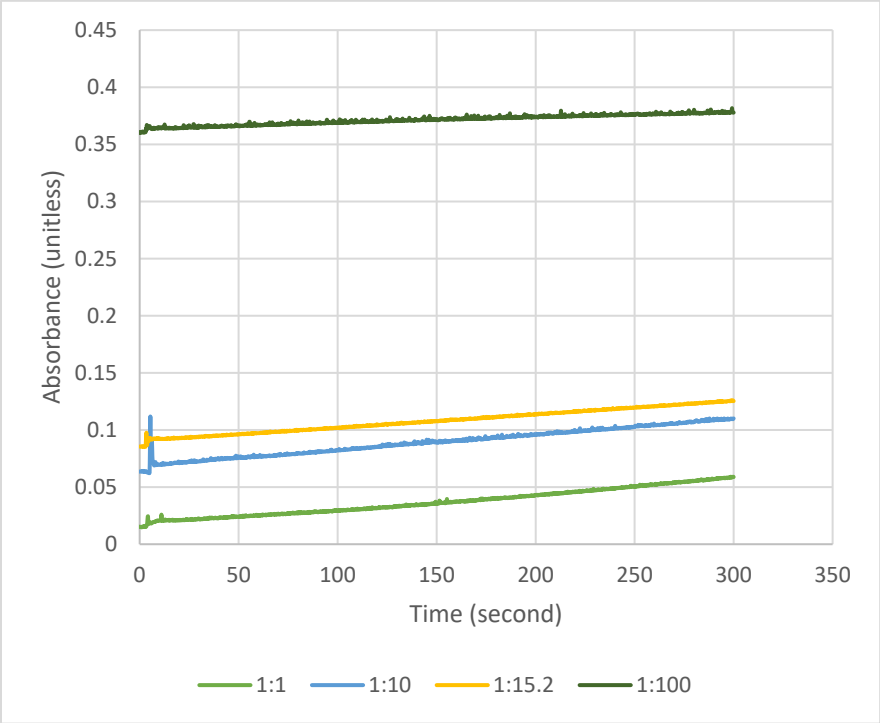

Figure 1: Catalytic Activity of GOx in Bound GOx-AuNP Systems. Each line is denoted by the ratio of GOx:HRP. None of the lines show indications of any reaction following enzymatic Michaelis-Menton kinetics, even as the favorability for the HRP to occur increases.

After successfully producing AuNP, the catalytic activity of the GOx at ratios GOx:HRP between 1:1 and 1:100. This was done so that an increasing ratio of HRP would favor the measurable proxy reaction over any other possible reaction between the hydrogen peroxide and the nanoparticle. As seen in Figure 1, all ratios showed no reaction occurring. This indicates that using the GOx as a reductive and stabilizing element for the nanoparticle formation causes the active site of the enzyme to become inactive. This could be caused by the active site being the source of the reduction or the formation of the nanoparticles causes a confirmation change in the enzyme that does not allow the substrate access. The cause of the inactivation is future direction for the project.

Additionally, we will synthesize GOx-bound AuPd nanoparticles using both synthesis methods. The first method will be to utilize the GOx to reduce and stabilize the AuPd nanoparticles. The second method will be to reduce the AuPd on its own with $NaBH_4$ and then bind the GOx to the AuPd nanoparticle surface. The catalytic activities of these two materials will be then compared. In addition to applications in catalysis, these enzyme-bound nanoparticles have broad applications in sensing and in medicine.