Enzymes in a golden cage

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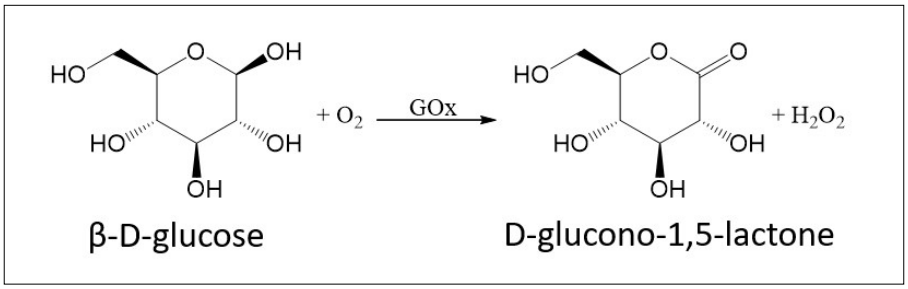
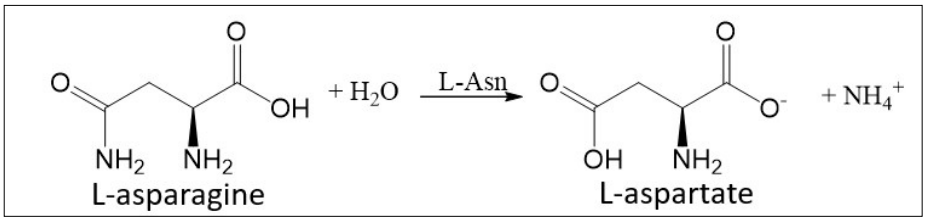
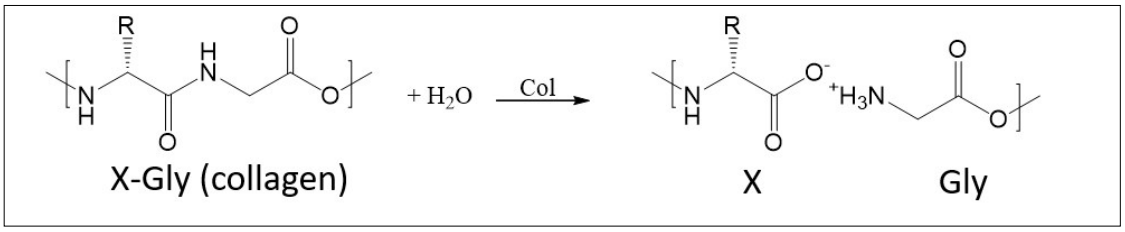
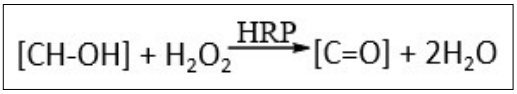
Supplementary Material

1. The enzymatic reactions of this study









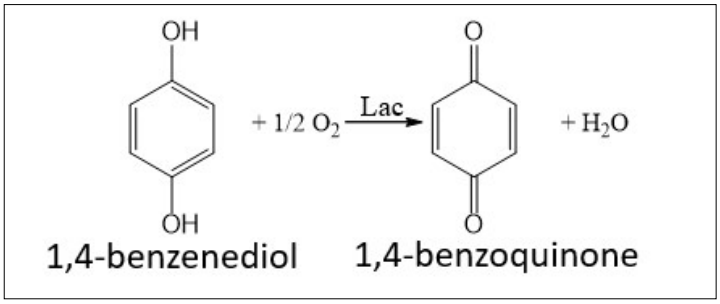
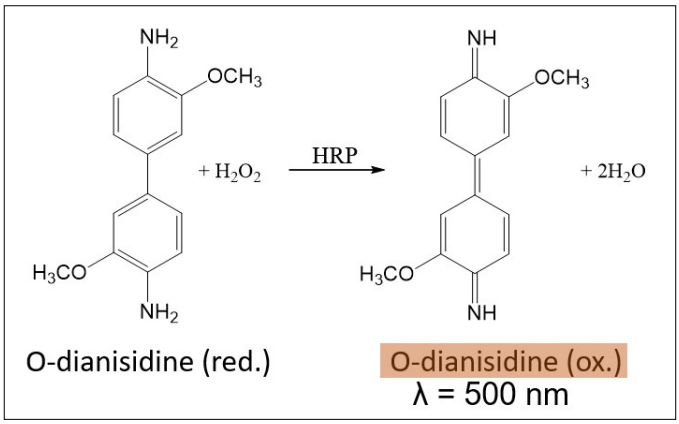
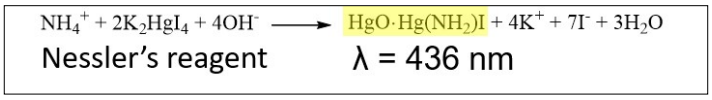
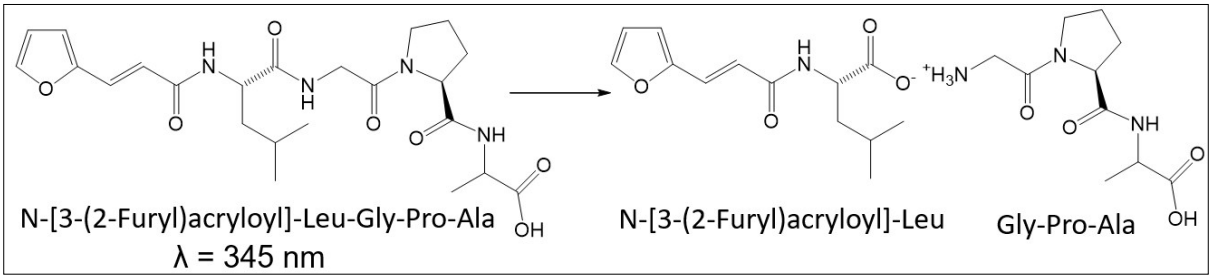
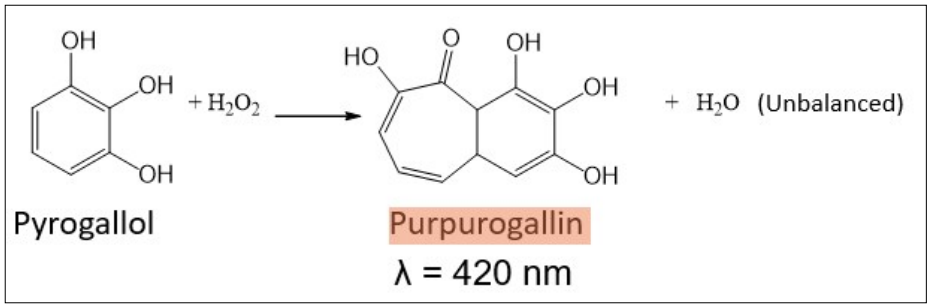


Figure S1 – Bio-reactions of each of the entrapped enzymes. From top to bottom: GOx, L-Asp, Col, HRP and Lac.









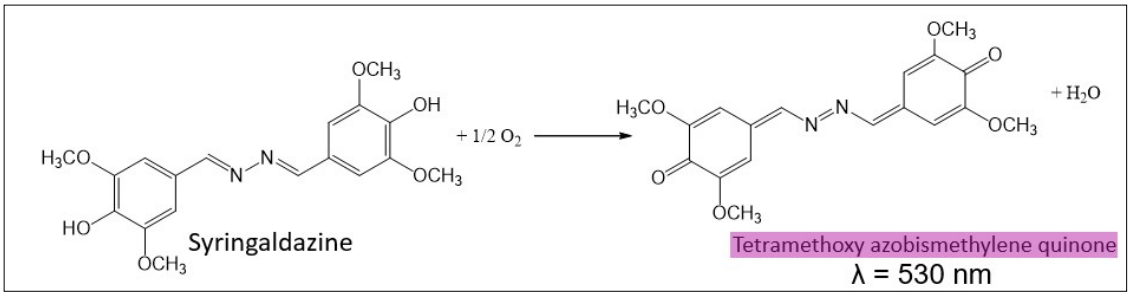


Figure S2 – The analytical reactions for the detection of each of the activity of each of the entrapped enzymes. From top to bottom: GOx, L-Asp, Col, HRP and Lac.

2. Surface maps of proteins

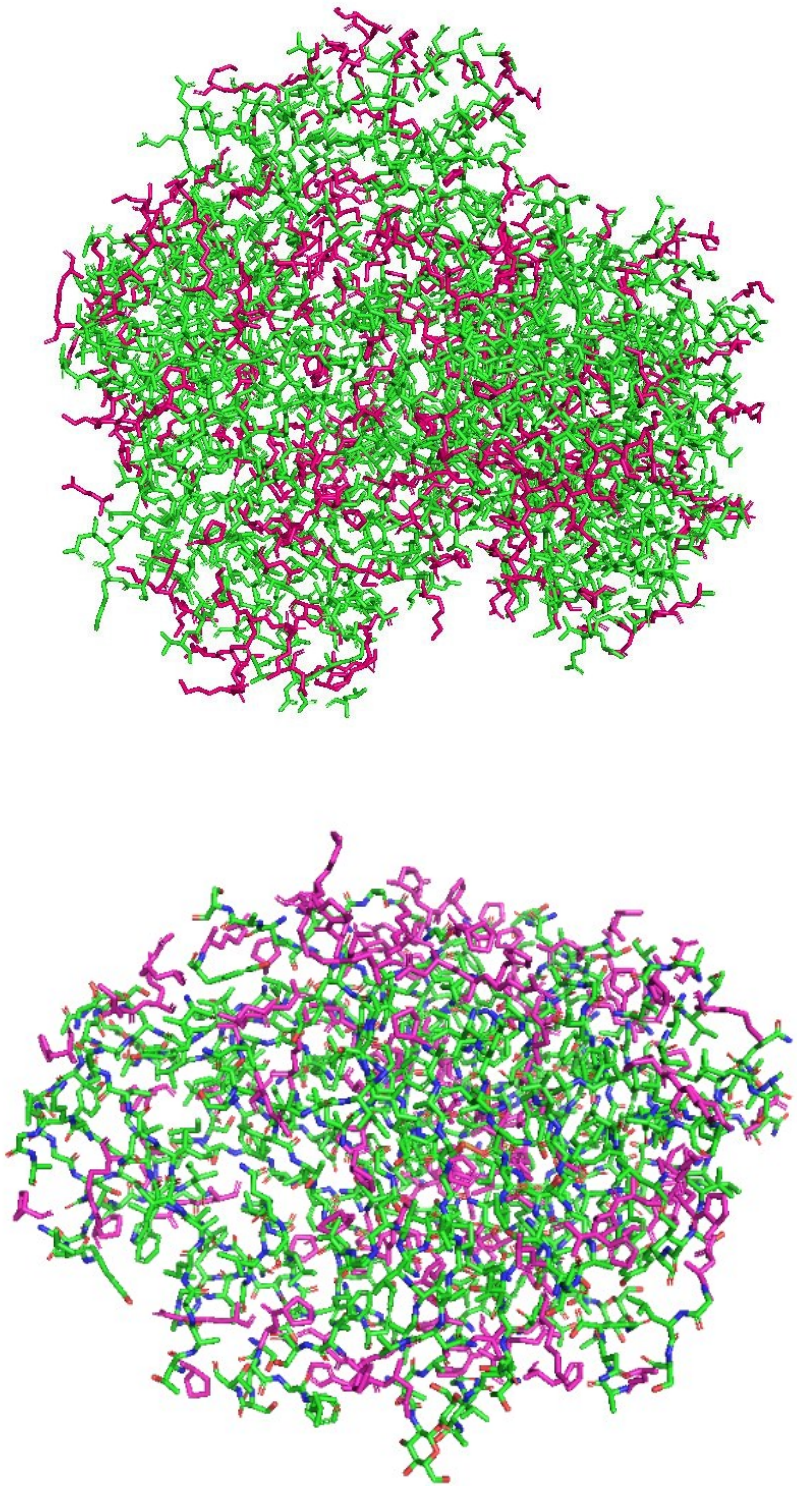


Figure S3 – Surface maps of L-asparaginase (top, PDB 6EOK) and laccase (bottom, PDB 1GYC): the residues with thiols, amines and carboxylates are indicated in pink

3. Additional materials characterization data

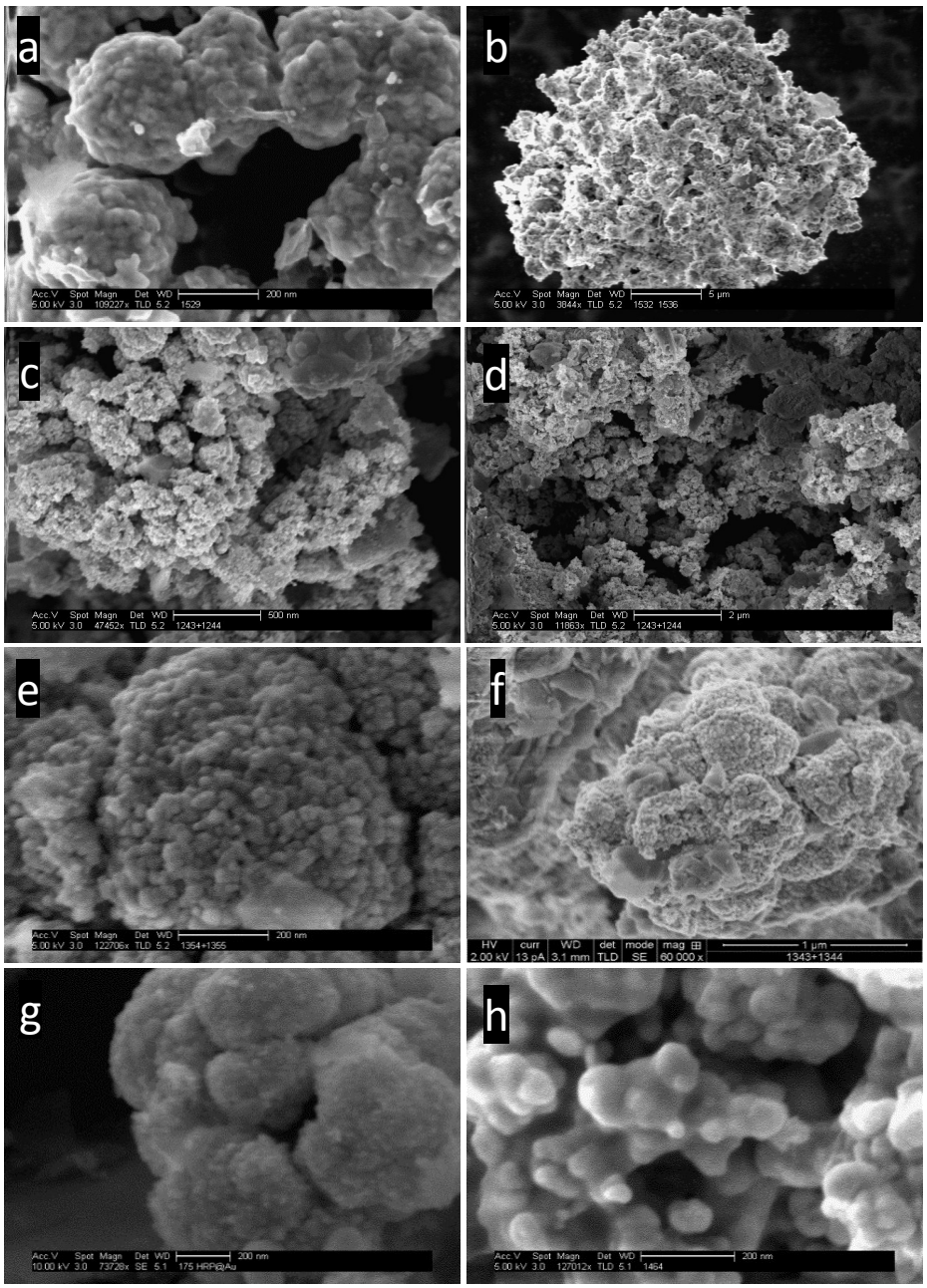


Figure S4 – HR-SEM images: GOx@Au: (a) bar = 200 nm; (b) bar = 5 µm. Asp@Au: (c) bar = 200 nm; (d) bar = 2 µm. (e) Col@Au: bar = 200 nm; (f) bar = 1 µm. (g) HRP@Au: bar = 200 nm. (h) Lac@Au: bar = 200 nm

Table S1 – Gold crystallite sizes of the composites from XRD

Composite	Average gold crystallite size
Asp@Au	14 nm
GOx@Au	16 nm
Col@Au	8 nm
Lac@Au	45 nm
Pure Gold	16 nm

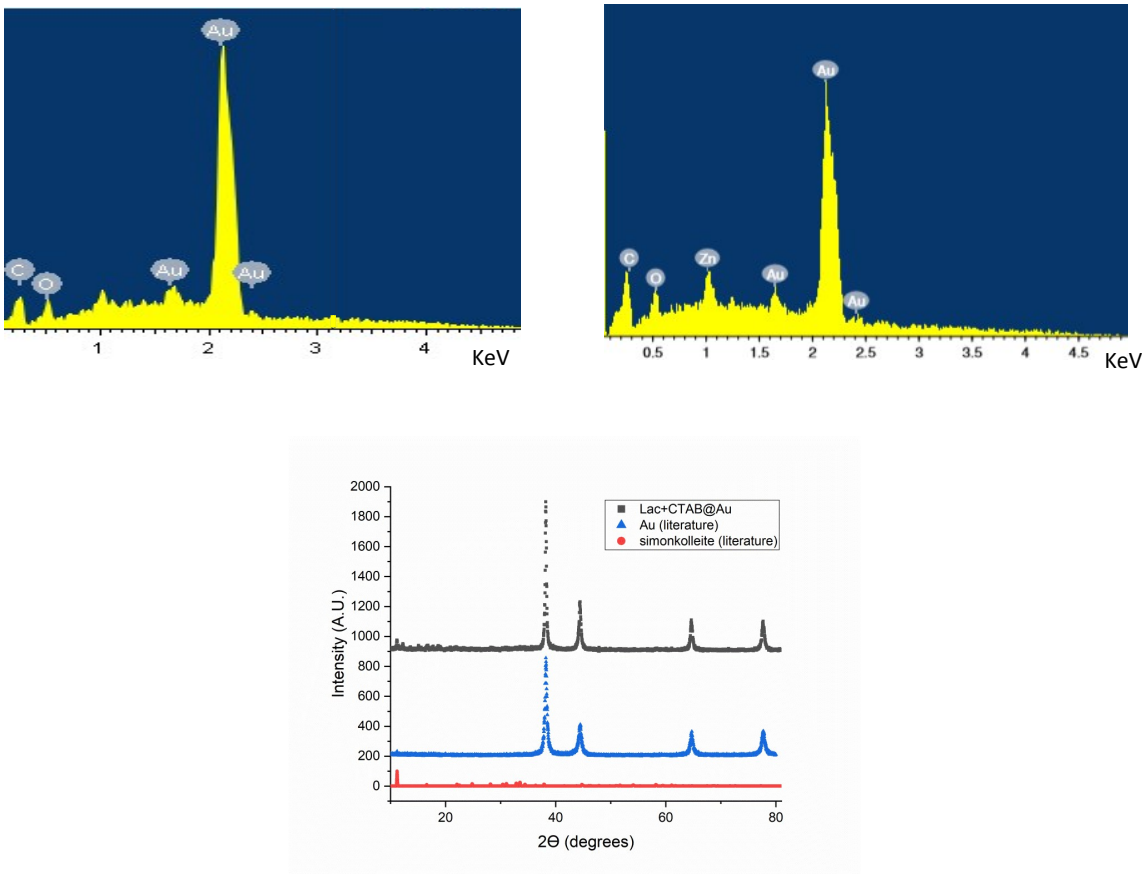


Figure S5 – Top: EDX spectra of Asp@Au (left) and Col@Au (right; residual traces of the reducing agent, $Zn(OH)_2$ are seen). Bottom: XRD of Lac@Au (black), Au (literature, blue). Traces of simonkolleite (literature, red) are seen.

3. Glucose oxidase kinetics in solution

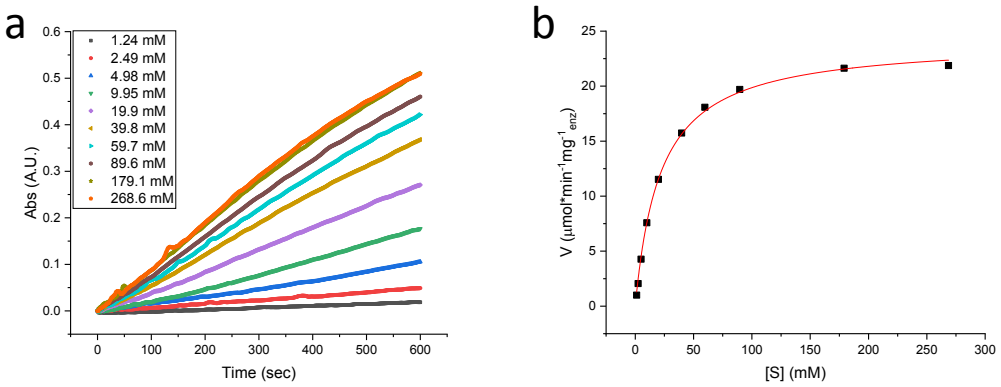


Figure S6 - Kinetics of free GOx in solution: (a) Initial rates as a function of the concentration of the substrate. (b) Fitting this data to the Michaelis-Menten model.

4. Collagenase: Additional observations

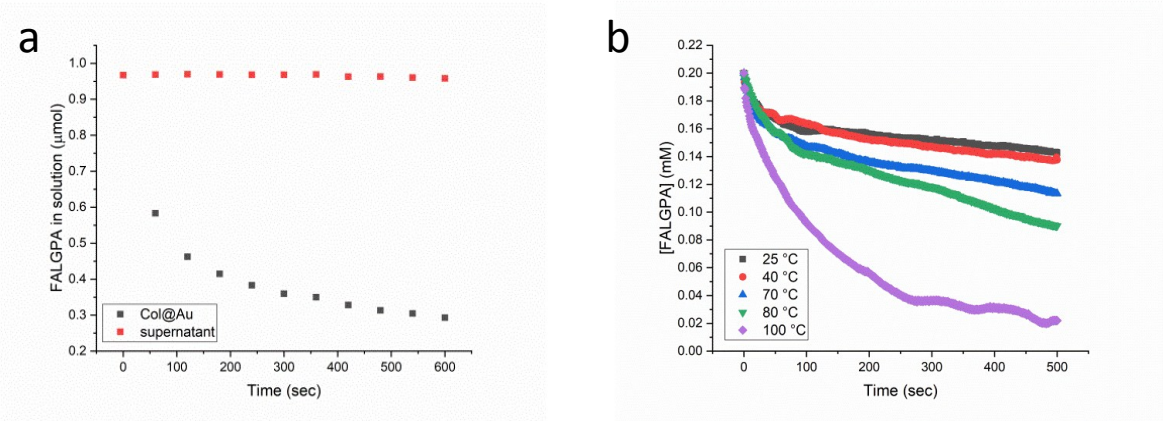


Figure S7 – (a) Activity of Col@Au, and zero activity of the supernatant solution. (b) Activity after heating the dry powder of Col@Au to the indicated temperature. We attribute the increase at 100 °C to pore widening due to coalescence of the Au nanocrystal building blocks.

5. Asparaginase – additional experiments

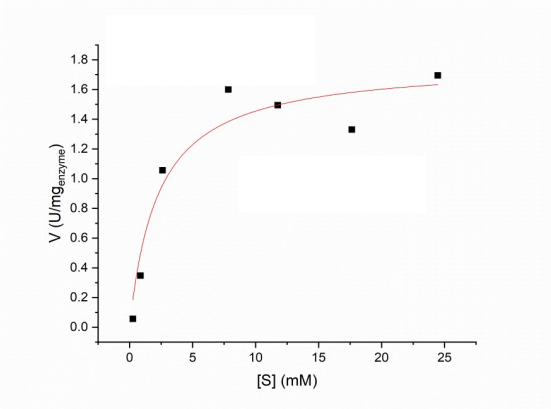


Figure S8 – Michaelis-Menten analysis of free L-Asp in solution

Testing for supernatant activity of Asp@Au was conducted in a different manner than for the composite, because even if enzyme leaked, it couldn't possibly stay active under the highly alkaline environment caused by Nessler's reagent. In order to establish that there was not enzyme leaking from the, it was soaked in 1.5 mL TDW for 30 minutes. The water was then measured for absorbance (in order to establish a blank measurement) and subsequently mixed with L-asparagine, the substrate for the enzyme, for 30 seconds, before injecting the Nessler's reagent and measuring the ensuing absorbance. The following change in absorbance was minute and at a base line level: from 0.0015 to 0.0029.