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Supporting Information

Sustainable Recycling of Formic Acid by Bio-Catalytic CO_2 Capture and Re-Hydrogenation

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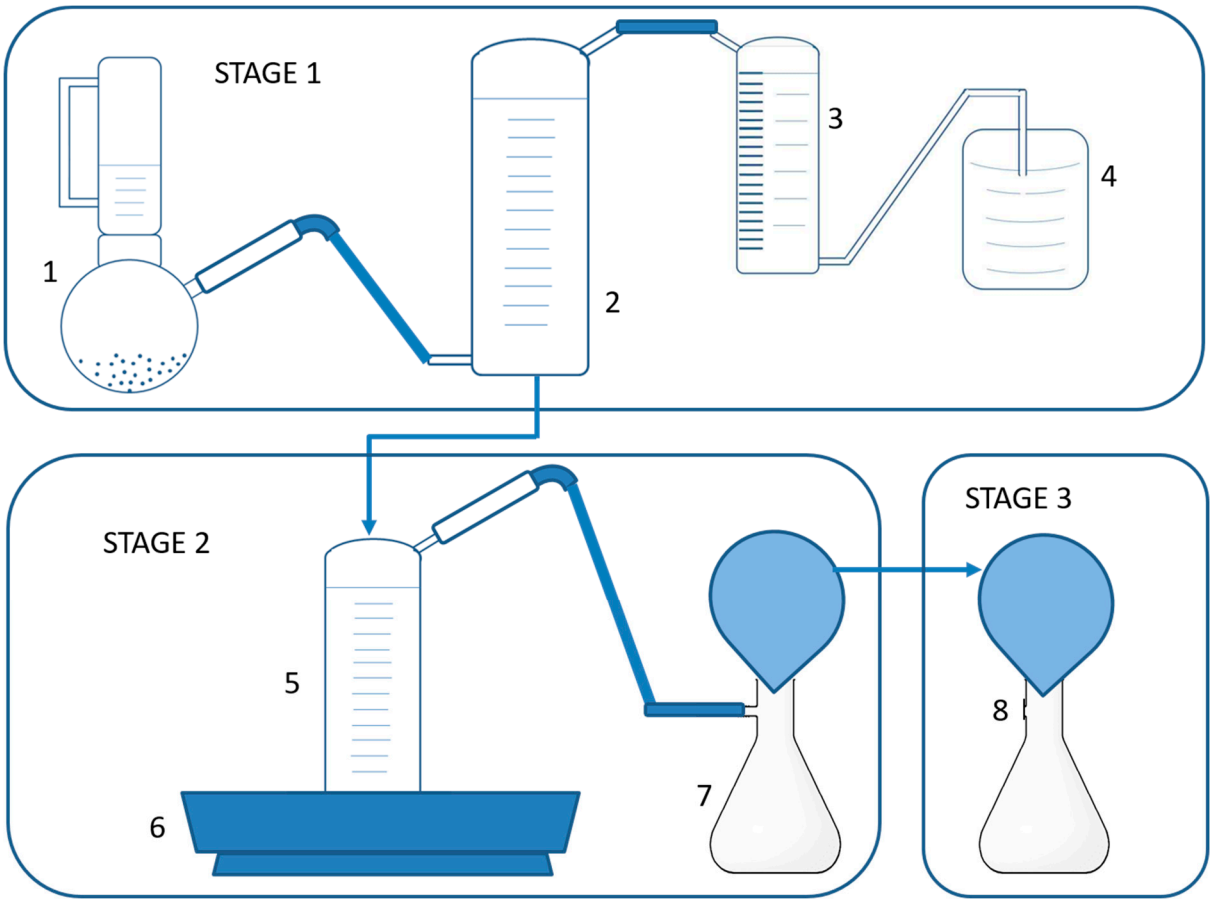

Figure S1. Schematic representation of laboratory set-up of the three stages in FA recycling process. Stage 1: FA decomposition and CO_2 capture, Stage 2: CO_2 stripping and Stage 3: CO_2 hydrogenation. Legends: 1-FA decomposition reaction flask containing Pd/C catalyst, 2-Buffer tank for CO_2 capture containing CA

enzyme/capture buffers, 3-graduated collection tank for H_2 and residual CO_2 gases, 4-Tank to collect water pushed out by the gases from the collection tank, 5-CO_2 stripping vessel, 6-Water bath, 7-Collection buffer flask with balloon connected, 8-CO_2 hydrogenation flask with FDH enzyme and NADH.

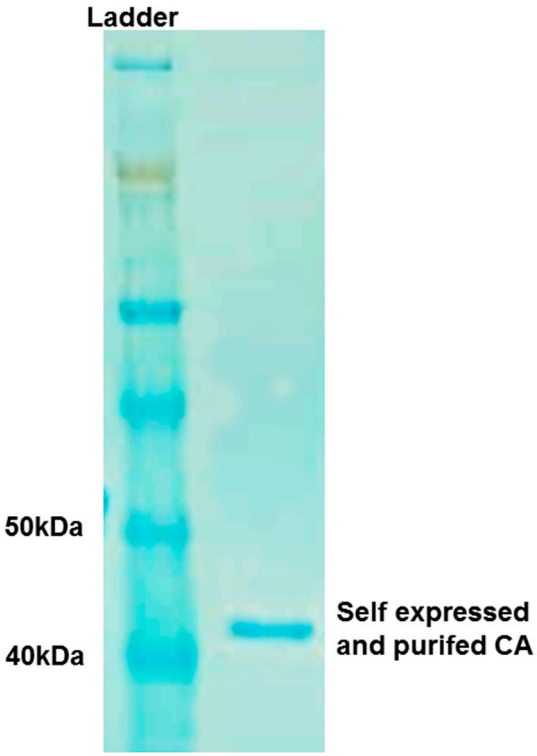

Figure S2. SDS-PAGE gel showing purified bovine carbonic anhydrase enzyme.

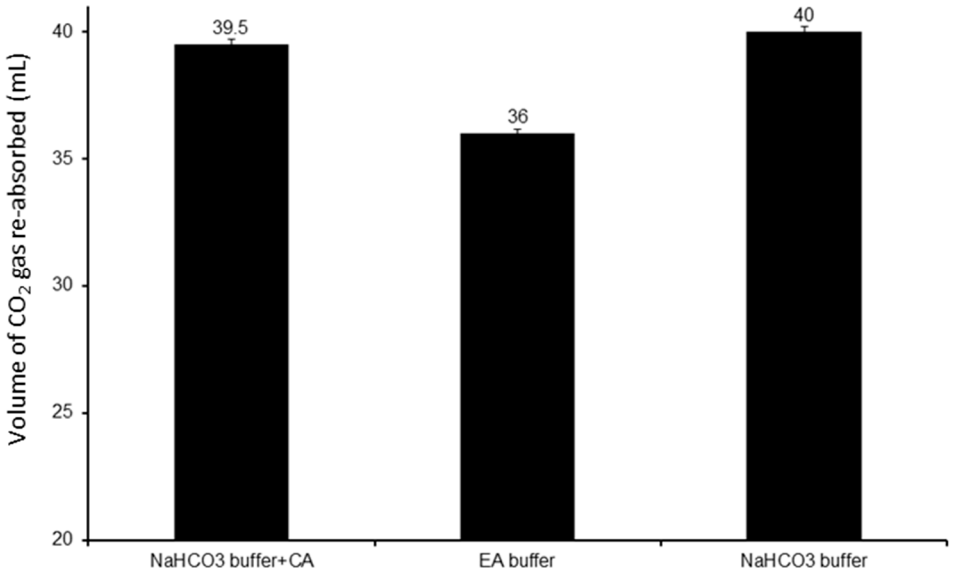

Figure S3. Reusability of stripped buffers presented by the volume of CO_2 absorbed by the capture buffers after being subjected to one stripping process.

CA enzyme heat treatment and activity assay

CA enzyme was subjected to heat treatment for 5 minutes at 50°C and 85°C using a digital dry bath heating block. CA enzyme at room temperature (25°C) without any heating was used as control. The enzyme activity was performed using *p*-nitrophenyl acetate as a substrate at a final concentration of 1mM in a 96 well assay plate in the reaction buffer containing 50mM sodium sulphate and 50 mM HEPES pH 8.0. A volume of 3µL of control and heat treated CA enzymes were added to the reaction mixture. The conversion of *p*-nitrophenyl acetate to *p*-nitrophenol is monitored by measuring the absorbance at 405 nm every 15 seconds for 15 minutes using microplate reader (Infinite 200PRO, Tecan). A blank solution was prepared using water instead of enzyme solution for absorbance correction. After blank correction of absorbance, the slope in terms of absorbance per second was calculated to determine reaction rate and relative activity of heat treated enzymes with respect to control.

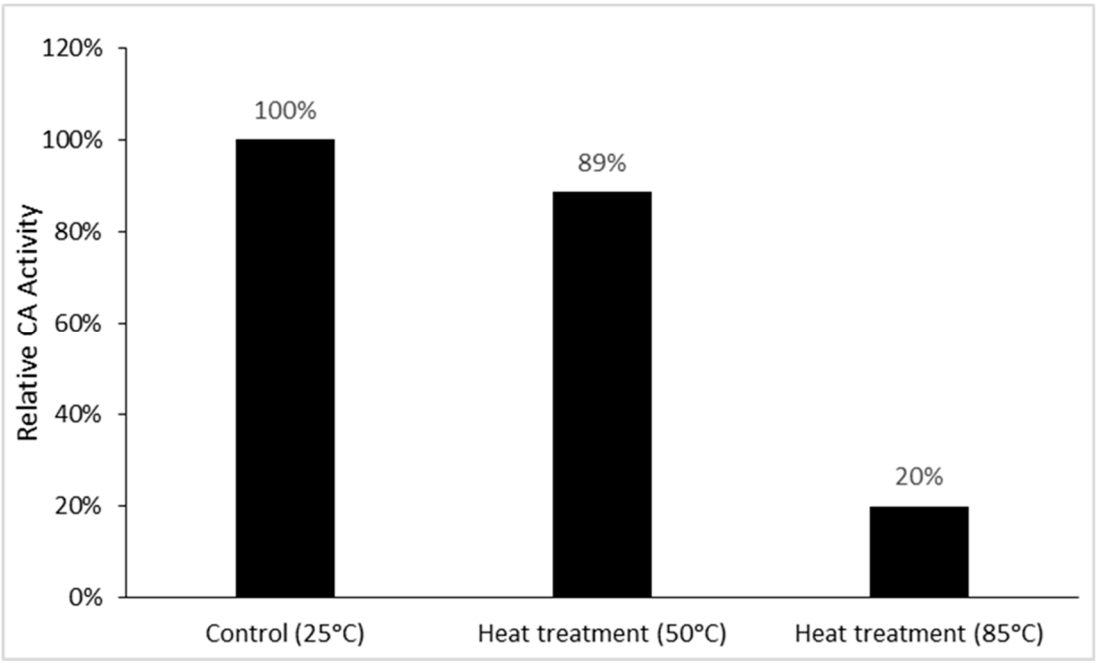

Figure S4. Relative activity of CA enzyme after heat treatment process.

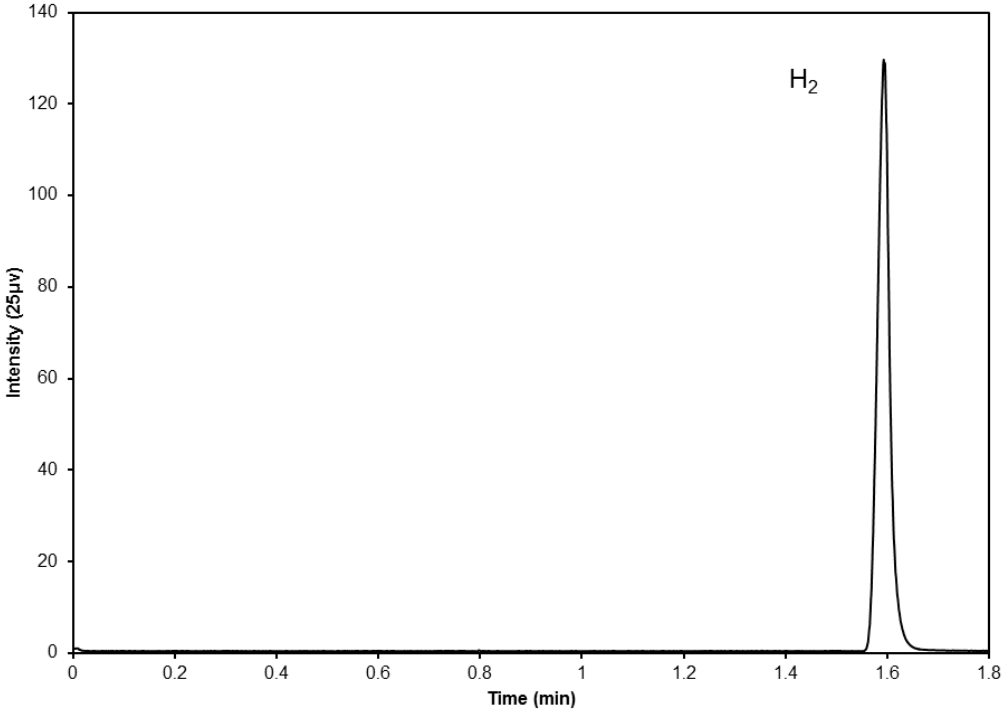

Figure S5. Gas chromatography profile showing the generation of H_2 gas with *in-situ* CO_2 capture using CA buffer

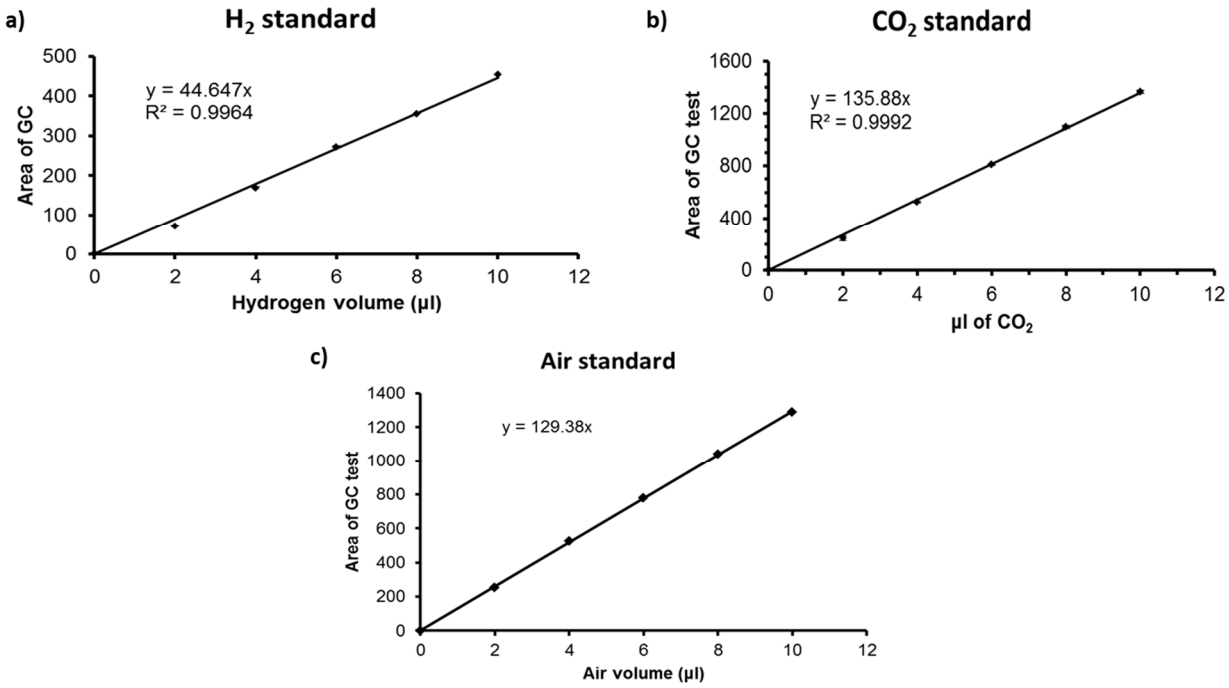

Figure S6. Standard curves for quantification of a) H_2, b) CO_2 and c) air measurements using gas chromatography.