**Electronic supporting information**

# Efficient Ionic Liquid–based platform for Multi-Enzymatic Conversion of Carbon Dioxide to Methanol

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**Synthesis of [Ch][AA] ILs.** ILs were prepared and purified according to the literature.[1] [Ch][OH] aqueous solution (about 4 M) was added dropwise under cooling to an amino acid aqueous solution or suspension to obtain a slight excess (about 10 mol%) of amino acid. Taking synthesis of [CH][Glu] as an example. 17.9 g of choline hydroxide (46 wt%) was diluted to 4 M with adding to 17 ml deionized water. Then prepared 4 M [Ch][OH] aqueous solution was added dropwise undercooling to 11g of L-glutamic acid dissolved in 200 ml deionized water ( about 0.4 M). The mixture was stirred at about 3 $^o$C overnight in the dark. Water was then removed under reduced pressure at 50 $^o$C using a rotavapor. Acetonitrile/methanol (9 : 1, v/v) was then added under vigorous stirring to precipitate the excess of amino acid. The mixture was left stirring overnight and the excess of amino acid was then filtered off. The filtrate was evaporated to remove solvents at 50 $^o$C. The product was dried under vacuum for 72 h at 60 $^o$C. The other synthesis of ILs was conducted with same procedure. All the peaks and corresponding chemical shifts obtained confirmed the structure of ILs. From NMR spectra and elemental analysis, the purity of ILs was more than 98 %.

**[CH][Glu]:** $^1$H NMR (600 MHz, $D_2O$, 25℃): δ = 2.04 – 2.23 (m, 2H, $CH_2$), 2.40 (apparent q, 2H, $CH_2$), 3.25 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.58 (apparent t, 2H, $CH_2$), 3.8 (q, J = 4.8, 7.2 Hz, 1H, CH-N), 4.11 (m, 2H, $CH_2$); $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ = 181.2, 174.5, 67.4, 55.6, 54.7, 53.8, 33.5, 26.9 ppm. Elemental analysis (%) calcd for: C 47.99, H 8.86, N 11.19; found C 47.96, H 8.62, N 11.46.

**[CH][Gly]:** $^1$H NMR (600 MHz, $D_2O$, 25℃): δ = 3.21 (s, 2H, $CH_2$-N), 3.24 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.54 –3.58 (m, 2H, $CH_2$), 4.10 (m, 2H, $CH_2$); $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ =180.5, 67.4, 55.6, 53.8, 33.5, 44.3 ppm. Elemental analysis (%) calcd for: C 47.17, H 10.18, N 15.72; found: C 47.05, H 10.50, N 15.81.

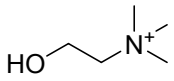
**[CH][His]:** $^1$H NMR (600 MHz, $D_2O$, 25℃): δ = 2.86 (m, 1H, $CH_2$), 2.97 (m, 1H, $CH_2$), 3.23 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.54 (apparent q, 3H, $CH_2$, CH-N), 4.07– 4.13 (m, 2H, $CH_2$), 6.96 (s, 1H, =CH), 7.71 (s, 1H, =CH); $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ =181.7, 135.8, 67.4, 56.0, 55.6, 53.8, 31.7 ppm. Elemental analysis (%) calcd for: C 51.15, H 8.58, N 21.69; found: C 51.02, H 8.67, N 21.75.

**[CH][Pro]:** $^1$H NMR (600 MHz, $D_2O$, 25℃): δ = 1.75 – 1.89 (m, 3H, $CH_2$, $CH_2$), 2.13 – 2.24 (m, 1H, $CH_2$), 2.84 – 2.93 (m, 1H, $CH_2$-N), 3.11 – 3.18 (m, 1H, $CH_2$-N), 3.24 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.56 – 3.73 (m, 3H, CH-N, $CH_2$), 4.07 – 4.13 (m, 2H, $CH_2$);

$^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ =180.6, 67.4, 61.4, 55.6, 53.8, 46.0, 30.3, 24.8 ppm; Elemental analysis (%) calcd for: C 55.02, H 10.16, N 12.83; found: C 53.97, H 9.71, N 12.28.

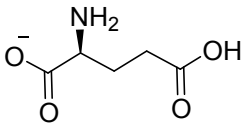
## 1. Structure of the cation and anions of [CH][AA] ILs.

**Cation**

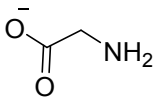


(Choline / CH)

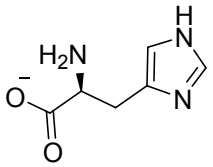
**Anions**



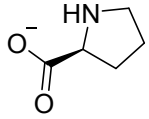
Glutamate / Glu



Glycine / Gly

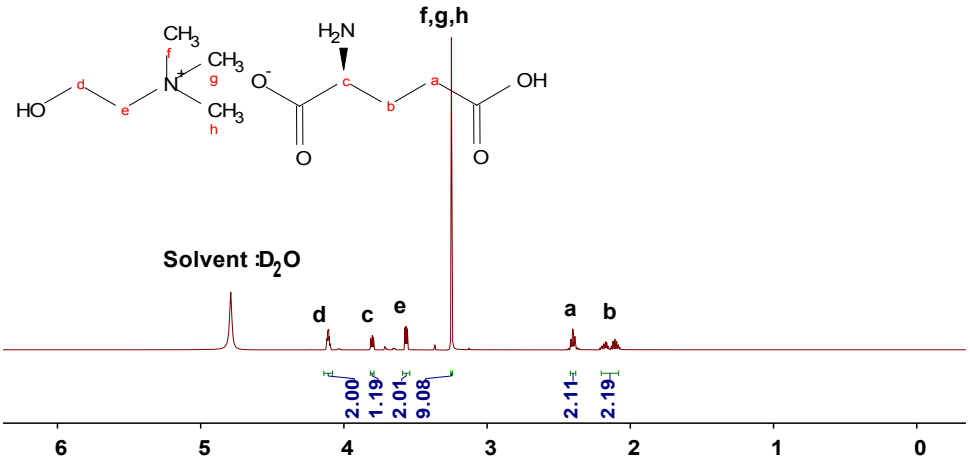


Histidine / His

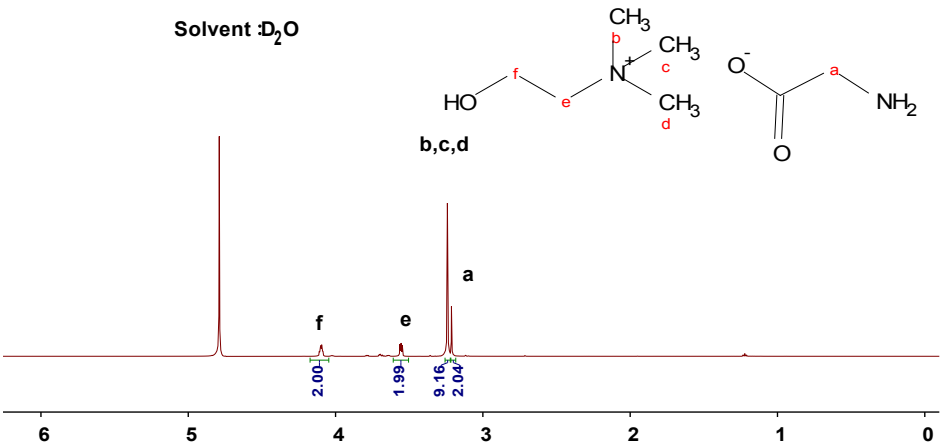


Proline / Pro

## 2. $^1$H NMR spectra for amino acid ionic liquids.




**[CH][Glu]:** $^1$H NMR (600 MHz, $D_2O$) δ: 2.04 – 2.23 (m, 2H, $CH_2$), 2.40 (apparent q, 2H, $CH_2$), 3.25 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.58(apparent t, 2H, $CH_2$), 3.8 (q, J = 4.8, 7.2 Hz, 1H, CH-N), 4.11 (m, 2H, $CH_2$).

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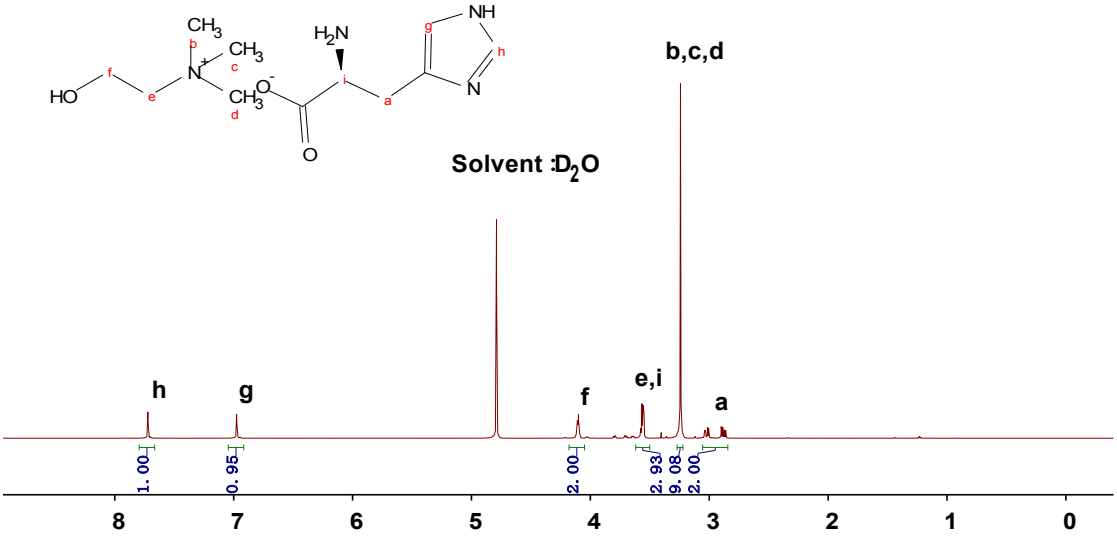
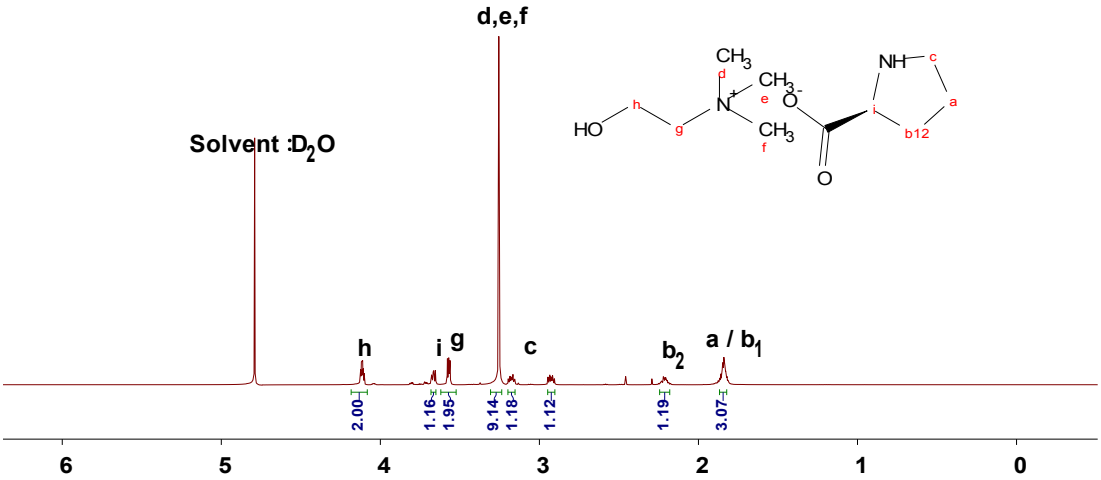
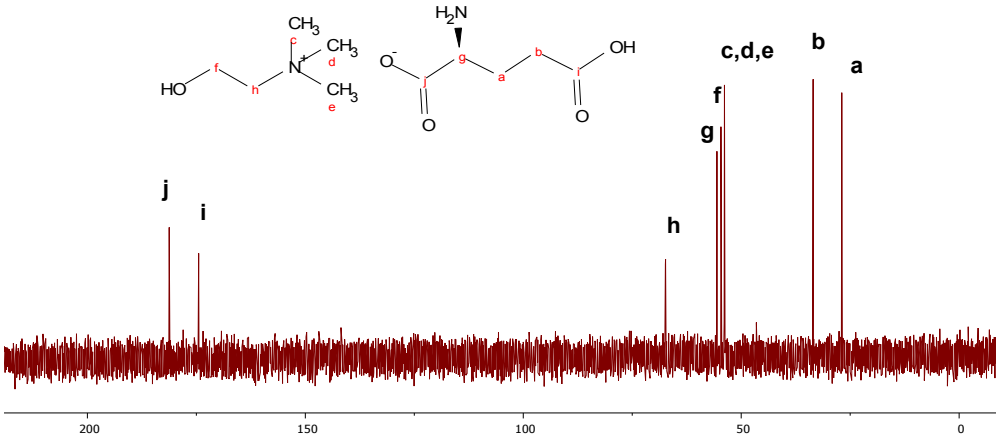



**[CH][His]:** $^1$H NMR (600 MHz, $D_2O$) δ: 2.86(m, 1H, $CH_2$), 2.97 (m, 1H, $CH_2$), 3.23 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.54 (apparent q, 3H, $CH_2$, CH-N), 4.07– 4.13 (m, 2H, $CH_2$), 6.96 (s, 1H, =CH), 7.71 (s, 1H, =CH).

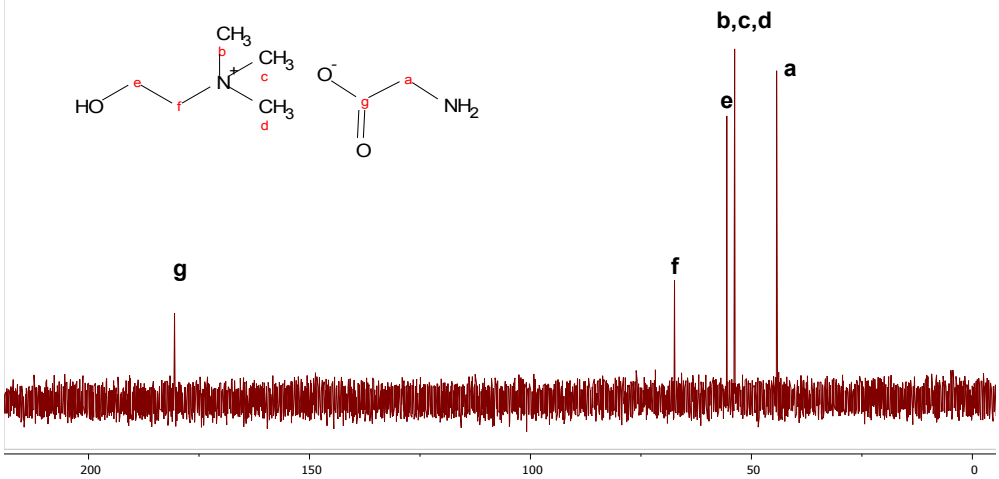



**[CH][Pro]:** $^{1}$H NMR (600 MHz, $D_2O$) δ: 1.75 – 1.89 (m, 3H, $CH_2$, $CH_2$), 2.13 – 2.24 (m, 1H, $CH_2$),2.84 – 2.93 (m, 1H, $CH_2$-N), 3.11 – 3.18 (m, 1H, $CH_2$-N), 3.24 (s, 9H, $CH_3$, $CH_3$, $CH_3$), 3.56 – 3.73 (m, 3H, CH-N, $CH_2$), 4.07 – 4.13 (m, 2H, $CH_2$).

## 3. $^{13}$C NMR spectra for amino acid ionic liquids.




**[CH][Glu]:** $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ=181.2, 174.5, 67.4, 55.6, 54.7, 53.8, 33.5, 26.9 ppm.



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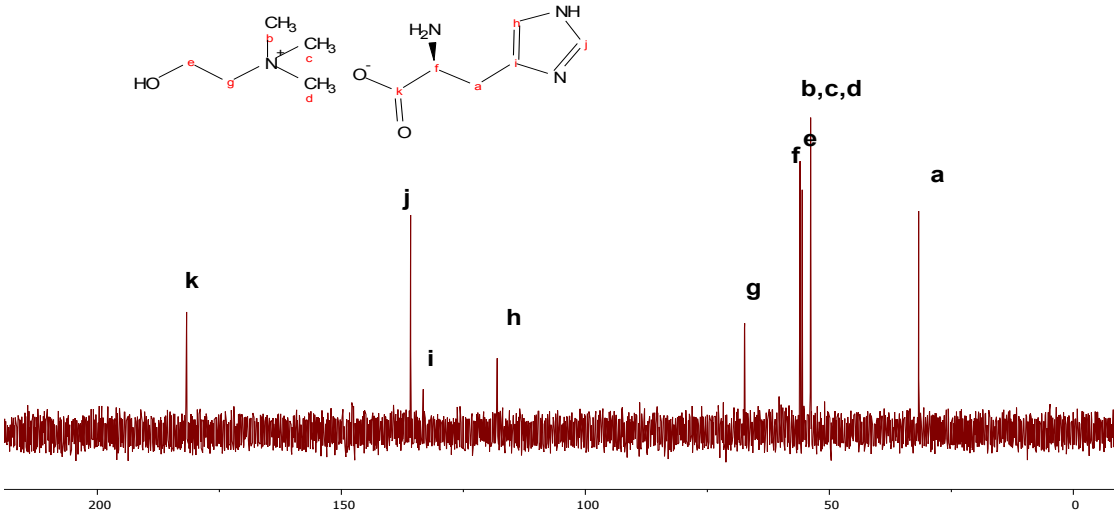
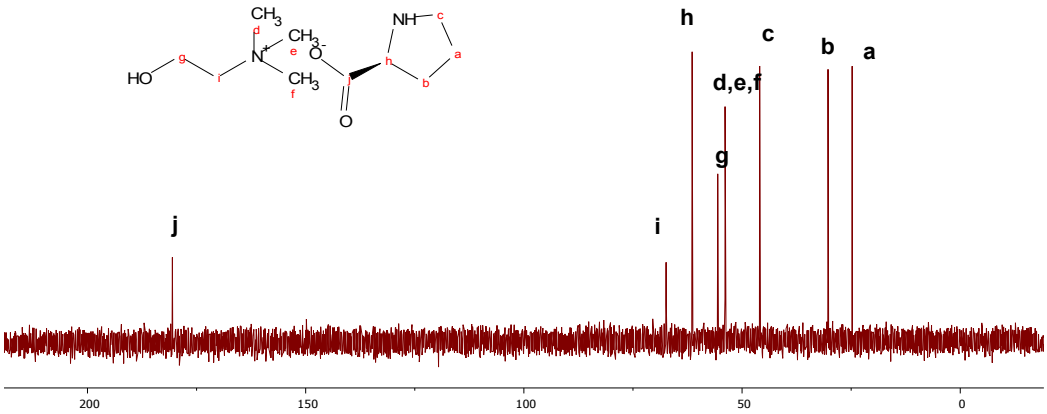



**[CH][His]:** $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ=181.7, 135.8, 67.4, 56.0, 55.6, 53.8, 31.7 ppm.




**[CH][Pro]:** $^{13}$C NMR (600 MHz, $D_2O$, 25℃) δ=180.6, 67.4, 61.4, 55.6, 53.8, 46.0, 30.3, 24.8 ppm.

## 4. Figures and Tables for results part.

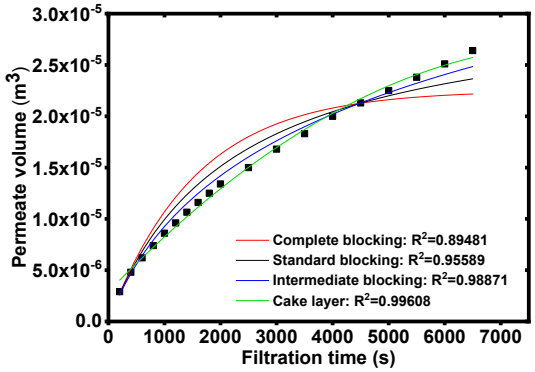



**Figure S1**. Evaluation of the predominant membrane fouling mechanisms at a constant pressure of 2 bar during immobilization.

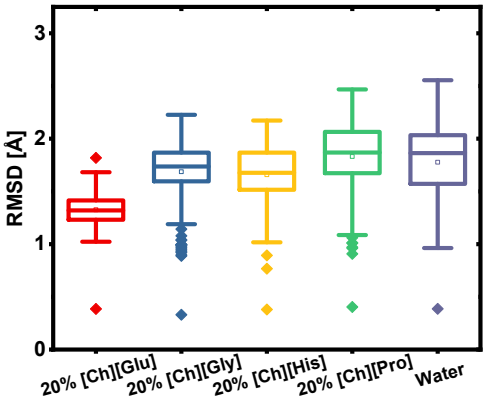



**Figure S2**: Statistical evaluation of the RMSD values for the Cα backbone over an MD simulation time of 18.1 ns.

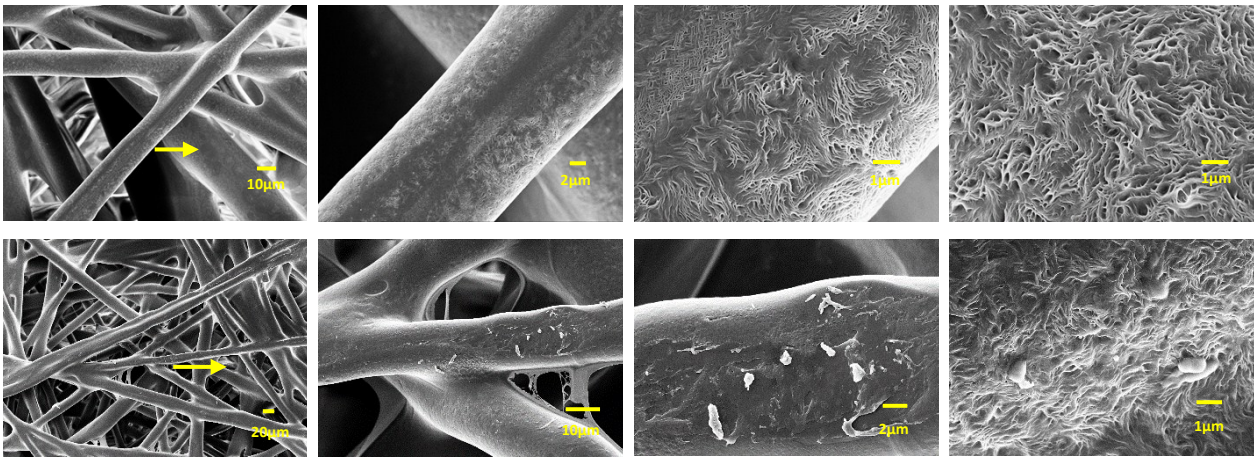



**Figure S3.** SEM images of Support layer (nonwoven polypropylene) before enzymes immobilization in the first line, and view of support layer after enzymes immobilization in the second line.

**Table S1.** Summary of membrane fouling models and mechanism under constant pressure filtration.

| Models | Equations | Schematic description |
|---|---|---|
| Complete blocking | $V = \frac{V_0}{k_b}[1 - exp(-k_b t)]$ |  |
| Standard blocking | $V = (V_0 t)(1 + \frac{V_0 k_s}{A_0} t)^{-1}$ |  |
| Intermediate blocking | $V = \frac{1}{k_i} ln(1 + k_i V_0 t)$ |  |
| Cake layer | $V = \frac{1}{V_0 k_c}(\sqrt{1 + (2 k_c V_0^2 t)} - 1)$ |  |

**Nomenclature**

| | | | |
|---|---|---|---|
| V | filtrate volume, $m^3$ | $k_b$ | complete blocking constant, $s^{-1}$ |
| t | filtration time, s | $k_s$ | standard blocking constant, $m^{-1}$ |
| $V_0$ | initial volumetric flowrate, $m^3/s$ | $k_i$ | intermediate blocking constant,$m^3$ |
| $A_0$ | initial membrane frontal area, $m^2$ | $k_c$ | cake filtration blocking constant, s $m^{-6}$ |

**Table S2.** Thermodynamic parameters determined.

| Enzyme | Ligand | Solvent | ΔG (KJ/mol) |
|---|---|---|---|
| FDH | $CO_2$ | Water | -17.12 |
| | $CO_2$ | 20%[CH][Glu]+water | -18.75 |

**Table S3.** Densities used for the molecular dynamics simulations

| Solvent | $\rho$ [g/cm$^3$] | Weighted $\rho$ [g/ cm$^3$] |
|---|---|---|
| $H_2O$ | 0,997 | 0,7976 |
| [Ch][Glu] | 1,168 | 0,2336 |
| [Ch][Gly] | 1,156 | 0,2312 |
| [Ch][His] | 1,204 | 0,2408 |
| [Ch][Pro] | 1,138 | 0,2276 |

**Table S4.** Detailed analysis of distances between residues and amino acid residues involved in formation of the transition state. All numbers are given in Å.

| Analyzed distances | Solvent system | | | | | |
|---|---|---|---|---|---|---|
| | 20% [Ch][Glu] | 20% [Ch][Gly] | 20% [Ch][His] | 20% [Ch][Pro] | $H_2O$ | TS[a] |
| C4(NAD)-C(CO2) | 3,596 ± 0,181 | 3,725 ± 0,251 | 4,069 ± 0,369 | 4,624 ± 0,505 | 17,990 ± 23,227 | 2,662 ± 0,064 |
| Ht-C4(NAD) | 1,085 ± 0,029 | 1,086 ± 0,029 | 1,089 ± 0,029 | 1,092 ± 0,028 | 1,088 ± 0,027 | 1,415 ± 0,032 |
| Ht-C(CO2) | 2,821 ± 0,182 | 3,034 ± 0,336 | 3,221 ± 0,412 | 3,880 ± 0,571 | 17,273 ± 23,271 | 1,304 ± 0,031 |
| O8(NAD)-C7(NAD) | 1,221 ± 0,022 | 1,224 ± 0,023 | 1,223 ± 0,021 | 1,220 ± 0,023 | 1,224 ± 0,023 | 1,266 ± 0,018 |
| C7(NAD)-C3(NAD) | 1,414 ± 0,024 | 1,421 ± 0,023 | 1,419 ± 0,025 | 1,419 ± 0,023 | 1,419 ± 0,023 | 1,491 ± 0,026 |
| O2(CO2)-HD21(N119) | 2,442 ± 0,386 | 4,271 ± 1,384 | 5,936 ± 1,910 | 7,029 ± 2,071 | 18,144 ± 22,442 | 1,978 ± 0,151 |

| | | | | | | |
|---|---|---|---|---|---|---|
| O2(CO2)-H(V93) | 2,440 ± 0,305 | 4,440 ± 1,388 | 5,364 ± 1,618 | 6,509 ± 1,802 | 17,635 ± 21,940 | 2,034 ± 0,158 |
| O1(CO2)-HH22(R258) | 2,916 ± 0,459 | 3,326 ± 0,972 | 3,914 ± 1,007 | 4,439 ± 1,073 | 16,764 ± 21,351 | 2,461 ± 0,293 |
| O1(CO2)-HH12(R258) | 2,913 ± 0,489 | 3,070 ± 0,882 | 3,593 ± 0,917 | 3,732 ± 0,936 | 17,100 ± 21,522 | 2,048 ± 0,179 |
| O2(CO2)-HH22(R258) | 4,286 ± 0,337 | 3,786 ± 0,989 | 3,805 ± 0,925 | 4,259 ± 1,059 | 16,225 ± 21,709 | 3,770 ± 0,207 |
| O2(CO2)-HH12(R258) | 4,824 ± 0,337 | 3,739 ± 0,976 | 3,636 ± 0,945 | 3,688 ± 0,929 | 16,346 ± 21,960 | 4,270 ± 0,189 |
| O1(CO2)-HE2(H311) | 3,009 ± 0,717 | 3,892 ± 0,806 | 4,638 ± 1,362 | 6,069 ± 1,854 | 18,338 ± 22,006 | 2,381 ± 0,250 |
| O8(NAD)-HE2(H311) | 2,767 ± 0,461 | 2,802 ± 0,397 | 6,581 ± 2,042 | 8,654 ± 2,067 | 3,484 ± 0,913 | 2,165 ± 0,212 |
| H92(NAD)-O(T256) | 2,983 ± 0,549 | 2,084 ± 0,178 | 2,141 ± 0,218 | 2,139 ± 0,261 | 2,460 ± 0,666 | 1,898 ± 0,146 |
| H91(NAD)-OD1(D282) | 2,515 ± 0,492 | 2,714 ± 0,364 | 3,131 ± 0,681 | 3,397 ± 0,456 | 3,411 ± 0,835 | 2,361 ± 0,231 |
| H91(NAD)-OD2(D282) | 2,952 ± 0,488 | 2,670 ± 0,455 | 4,907 ± 0,956 | 5,186 ± 0,784 | 3,816 ± 1,253 | 2,412 ± 0,295 |
| H91(NAD)-OG(S313) | 2,824 ± 0,355 | 2,696 ± 0,314 | 3,077 ± 0,757 | 2,960 ± 0,618 | 3,159 ± 0,756 | 2,721 ± 0,217 |
| HG(S313)-OD2(D282) | 2,805 ± 0,164 | 2,692 ± 0,133 | 3,930 ± 0,600 | 4,211 ± 0,464 | 3,163 ± 0,778 | 1,751 ± 0,155 |
| ND1(H311)-HE21(Q287) | 4,627 ± 0,358 | 3,327 ± 0,734 | 3,829 ± 0,527 | 5,922 ± 0,593 | 3,516 ± 0,735 | 3,442 ± 0,159 |
| ND1(H311)-HE22(Q287) | 5,775 ± 0,300 | 3,763 ± 0,934 | 4,657 ± 0,626 | 6,561 ± 0,961 | 4,067 ± 1,000 | 1,862 ± 0,120 |

[a] as reported by Castillo, et al. (J. Phys. Chem. B 2008, 112, 10012–10022)

[1] S. De Santis, G. Masci, F. Casciotta, R. Caminiti, E. Scarpellini, M. Campetella, L. Gontrani, *Phys Chem Chem Phys* **2015**, *17*, 20687-20698.