

Enzyme Immobilization on Stimuli-Responsive Hydrogels - Part A.

Influence of Carrier Properties on Immobilization Efficiency and Catalytic Performance

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This Supplementary Information file contains the numerical values of the data points presented in the main publication. This dataset is available through an external repository, accompanied by a descriptive README file: <https://doi.org/10.15480/882.16752>.

RESULTS

Table S 1. Degree of swelling and elastic modulus of hydrogels at 25 °C in distilled water.

Hydrogel	Degree of Swelling (-)	Elastic Modulus (kPa)
p(HEMA-co-IA)	4.57 ± 0.28	5.76 ± 0.79
p(NIPAM-co-IA)	85.37 ± 5.30	4.74 ± 0.53
pNIPAM	42.87 ± 3.36	1.39 ± 0.05

Table S 2. Relative swelling and shrinkage of hydrogels. Temperature-responsive swelling from 40 °C to 25 °C (in water) and pH-responsive swelling from pH 4 to pH 8 (phosphate buffer)

Hydrogel	Stimulus	Swelling Ratio (-)
p(HEMA-co-IA)	Temperature	1.00 ± 0.08
p(NIPAM-co-IA)	Temperature	0.81 ± 0.08
pNIPAM	Temperature	0.14 ± 0.04
p(HEMA-co-IA)	pH	0.57 ± 0.03
p(NIPAM-co-IA)	pH	0.63 ± 0.04
pNIPAM	pH	1.00 ± 0.04

Table S 3. Immobilization yield of FDH on hydrogel monoliths and particles for adsorptive binding (24 h), and covalent binding (3 h and 24 h).

Hydrogel	Shape	Binding Mechanism	Immobilization Yield (%)
p(HEMA-co-IA)	Monolith	Adsorption (24 h)	41.81 ± 14.08
p(HEMA-co-IA)	Monolith	Covalent Binding (3 h)	22.34 ± 2.20
p(HEMA-co-IA)	Monolith	Covalent Binding (24 h)	46.23 ± 4.20
p(NIPAM-co-IA)	Monolith	Adsorption (24 h)	34.02 ± 4.98
p(NIPAM-co-IA)	Monolith	Covalent Binding (3 h)	15.94 ± 5.28
p(NIPAM-co-IA)	Monolith	Covalent Binding (24 h)	38.99 ± 3.74
pNIPAM	Monolith	Adsorption (24 h)	32.40 ± 8.72
pNIPAM	Monolith	Covalent Binding (3 h)	14.40 ± 2.20
pNIPAM	Monolith	Covalent Binding (24 h)	32.02 ± 8.37
p(HEMA-co-IA)	Particles	Covalent Binding (24 h)	31.54 ± 5.45
p(NIPAM-co-IA)	Particles	Covalent Binding (24 h)	19.72 ± 7.88
pNIPAM	Particles	Covalent Binding (24 h)	10.69 ± 4.51
silica	Particles	Adsorption (24 h)	48.09 ± 6.07

Table S 4. Enzyme loading of FDH immobilized on hydrogel monoliths and particles using adsorptive binding (24 h), and covalent binding (3 h and 24 h).

Hydrogel	Shape	Binding Mechanism	Enzyme Loading (mg _{Enzyme} /g _{Polymer})
p(HEMA-co-IA)	Monolith	Adsorption (24 h)	2.15 ± 1.25
p(HEMA-co-IA)	Monolith	Covalent Binding (3 h)	1.29 ± 0.38
p(HEMA-co-IA)	Monolith	Covalent Binding (24 h)	2.26 ± 0.38
p(NIPAM-co-IA)	Monolith	Adsorption (24 h)	12.15 ± 1.78
p(NIPAM-co-IA)	Monolith	Covalent Binding (3 h)	7.63 ± 2.59
p(NIPAM-co-IA)	Monolith	Covalent Binding (24 h)	12.51 ± 1.38
pNIPAM	Monolith	Adsorption (24 h)	10.63 ± 1.78
pNIPAM	Monolith	Covalent Binding (3 h)	5.55 ± 1.88
pNIPAM	Monolith	Covalent Binding (24 h)	9.20 ± 2.33
p(HEMA-co-IA)	Particles	Covalent Binding (24 h)	1.55 ± 0.24
p(NIPAM-co-IA)	Particles	Covalent Binding (24 h)	5.06 ± 2.18
pNIPAM	Particles	Covalent Binding (24 h)	3.98 ± 1.70
silica	Particles	Adsorption (24 h)	0.76 ± 0.14

Table S 5. Activity yield of FDH immobilized on hydrogel monoliths using adsorptive binding (24 h), and covalent binding (3 h and 24 h).

Hydrogel	Shape	Binding Mechanism	Activity Yield (%)
p(HEMA-co-IA)	Monolith	Adsorption (24 h)	2.48 ± 1.15
p(HEMA-co-IA)	Monolith	Covalent Binding (3 h)	1.96 ± 0.60
p(HEMA-co-IA)	Monolith	Covalent Binding (24 h)	2.63 ± 0.71
p(NIPAM-co-IA)	Monolith	Adsorption (24 h)	1.61 ± 0.44
p(NIPAM-co-IA)	Monolith	Covalent Binding (3 h)	1.66 ± 0.35
p(NIPAM-co-IA)	Monolith	Covalent Binding (24 h)	2.17 ± 0.41
pNIPAM	Monolith	Adsorption (24 h)	2.69 ± 1.26
pNIPAM	Monolith	Covalent Binding (3 h)	1.87 ± 0.69
pNIPAM	Monolith	Covalent Binding (24 h)	0.98 ± 0.32

Table S 6. Relative leaching of FDH immobilized on hydrogel monoliths using adsorptive binding (24 h), and covalent binding (3 h and 24 h).

Hydrogel	Shape	Binding Mechanism	Relative Leaching (%)
p(HEMA-co-IA)	Monolith	Adsorption (24 h)	17.40 ± 10.37
p(HEMA-co-IA)	Monolith	Covalent Binding (3 h)	18.03 ± 4.74
p(HEMA-co-IA)	Monolith	Covalent Binding (24 h)	14.82 ± 2.93
p(NIPAM-co-IA)	Monolith	Adsorption (24 h)	30.92 ± 11.40
p(NIPAM-co-IA)	Monolith	Covalent Binding (3 h)	10.68 ± 1.53
p(NIPAM-co-IA)	Monolith	Covalent Binding (24 h)	6.81 ± 4.05
pNIPAM	Monolith	Adsorption (24 h)	33.99 ± 15.09
pNIPAM	Monolith	Covalent Binding (3 h)	27.08 ± 9.53
pNIPAM	Monolith	Covalent Binding (24 h)	14.70 ± 5.11