

Supporting Information for:

Precise, fast and flexible determination of protein interactions by affinity capillary electrophoresis: Part 3: Anions

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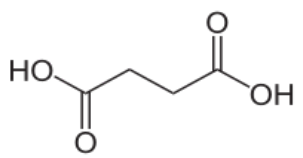
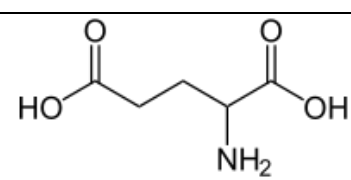
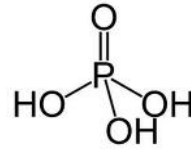
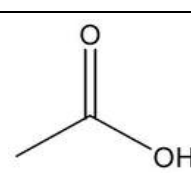
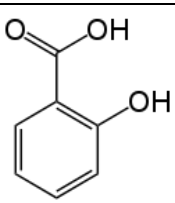
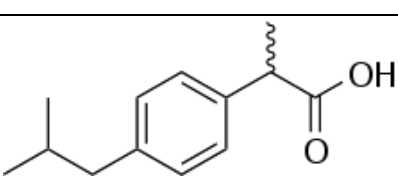
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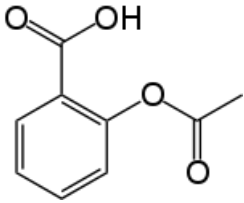
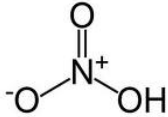
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Yuanhong Xu and Sabine Redweik contributed equally to this work.

Table S1. pK_a values and structures of the studied anions

Anion existing at the working pH 7.4	Corresponding acid form of the anion	pK _a	Structures
Succinate	Succinic acid	4.2 5.6 [S1]	
Glutamate	Glutamic acid	2.19 4.25 9.67 pI = 3.08 [S2]	
Phosphate	Phosphoric acid ^b	2.12 7.21 12.67 [S3]	
Acetate	Acetic acid ^b	4.76 [S4]	
Salicylate	Salicylic acid	2.97 [S5]	
Iso-phenylpropionate ^a	Ibuprofen	4.5 [S6]	

Acetyl salicylate ^a	Aspirin	3.5 [S7]	
Iodide	Hydroiodic acid ^b	-9.5 [S8]	$\text{H}-\text{I}$
Nitrate	Nitric acid ^b	-1.4 [S9]	
Thiocyanate	Thiocyanic acid ^b	1.1 [S10]	$\text{H}-\text{S}-\text{C}\equiv\text{N}$

Note: a, although the anions existed as the form of iso-phenylpropionate and acetyl salicylate for ibuprofen and aspirin at the working pH 7.4, respectively, “ibuprofen” and “aspirin” were actually used when mentioned in the main text given the purpose of easy description.

b, in our experiment, the corresponding salts were used, they are disodium hydrogen phosphate, sodium acetate, potassium iodide, sodium nitrate, potassium thiocyanate, respectively.

[S1] http://en.wikipedia.org/wiki/Succinic_acid.

[S2] http://www.anaspec.com/html/pK_n_pl_Values_of_AminoAcids.html.

[S3] http://en.wikipedia.org/wiki/Phosphoric_acid.

[S4] http://en.wikipedia.org/wiki/Acetic_acid.

[S5] http://en.wikipedia.org/wiki/Salicylic_acid.

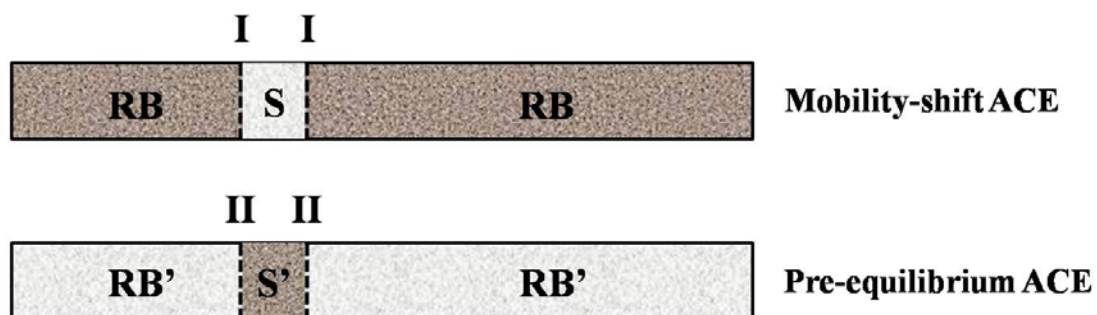
[S6] Itagaki, S., Gopal, E., Zhuang, L., Fei, Y.-J., Miyauchi, S., Prasad, P. D., Ganapathy, V., *Pharm. Res.* 2006, 23, 1209-1216.

[S7] <http://en.wikipedia.org/wiki/Aspirin>.

[S8] http://en.wikipedia.org/wiki/Hydrogen_iodide.

[S9] http://en.wikipedia.org/wiki/Nitric_acid.

[S10] http://en.wikipedia.org/wiki/Thiocyanic_acid.



S: protein; RB: running buffer with anion

S': preincubated protein-anion; RB': running buffer without anion

Fig. S1 Schematic diagram of equilibrium (above) and non-equilibrium ACE (below).

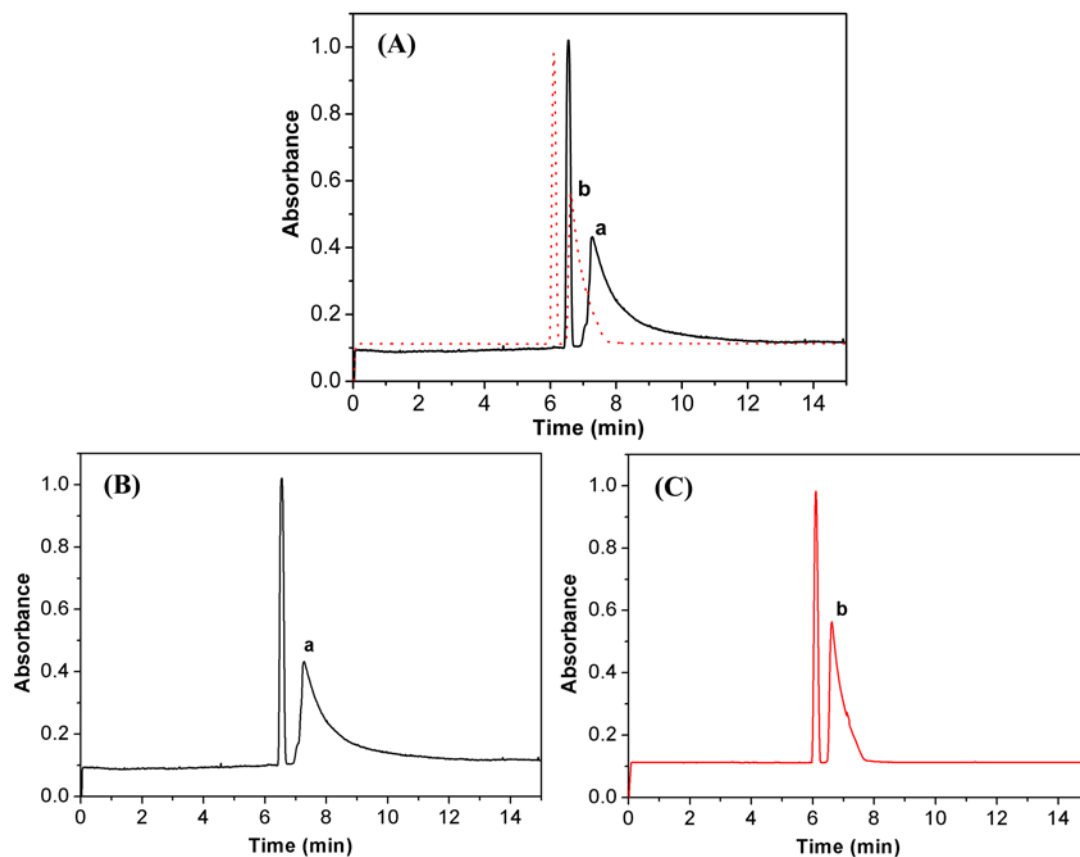


Fig. S2 Electrophoretic mobility changes of 50 μM myoglobin in the absence (a) and presence of (b) 500 μM phosphate using Tris running buffer by the equilibrium ACE method.

(A) Overlay curves of (a) and (b); separate curves of (B) (a) and (C) (b), respectively.

The left peak in each electrophoretic curve was the UV adsorption signal of the neutral marker acetanilide, the other one is that of myoglobin.

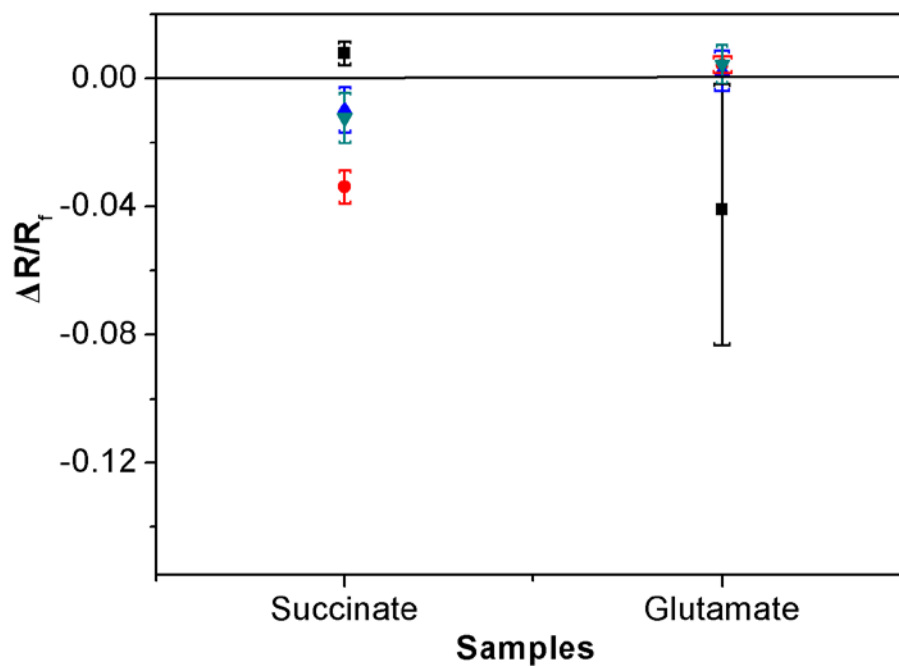


Fig. S3 Interactions using phosphate buffer, between the anions (succinate, or glutamate) and the proteins BSA (black \blacksquare), β -lactoglobulin (red \bullet), and ovalbumin (blue \bullet and green \bullet , two of the isoforms obviously indicated in the electropherogram) studied by equilibrium ACE method. ΔR is normalized to R_f and then the specific confidence intervals are shown.