Supplementary Material (ESI) for Analyst


# Review on Theory of Moving Reaction Boundary, Electromigration Reaction Methods and Applications in Isoelectric Focusing and Sample Pre-concentration

**Cheng-Xi Cao*[1], Liu-Yin Fan[1] and Wei Zhang[1]**

[1]*Laboratory of Analytical Biochemistry & Bio-separation, Key Laboratory of Microbiology of Educational Ministry, School of Life Science and Biotechnology, Shanghai Jiao Tong University, Shanghai 200240, China*

---

* The correspondence author: Professor Cheng-Xi Cao, School of Life Science and Biotechnology, Shanghai Jiao Tong University, 800 Dongchuan Rd., Shanghai 200240, P. R. China. email: cxcao@sjtu.edu.cn, Fax: 86-21-3420 5820, phone: 86-21-3420 5682

**Table S1**. Agreements between prediction of $R_r$ values and experiments on pH gradient drifting for different acid-base pairs in IEF in refs.[a)]

| No. | Run of IEF | Acid_base pairs | $R_r$ | Prediction [b)] | Experiment[b)] |
|---|---|---|---|---|---|
| 1 | Fig. 4 and 5 in Ref. [57] | 0.2% $H_2SO_4$<br>0.4% Ethylenediamine | 0.03 | + → –, w | + → – w |
| 2 | Fig. 1 and 3 in Ref. [60] | 100 mM $H_2SO_4$<br>200 mM KOH | 0.22 | + → –, o | + → – o |
| 3 | Fig. 1 in Ref. [62] | 100 mM $H_2SO_4$<br>200 mM KOH | 0.22 | + → –, o | + → –, o |
| 4 | Fig. 1, 3D and 4B in Ref. [64] | 100 mM $H_2SO_4$<br>200 mM KOH | 0.22 | + → –, o | + → –, o |
| 5 | Fig. 1B in Ref. [65] | 100 mM $H_2SO_4$<br>200 mM KOH | 0.22 | + → –, o | + → –, o |
| 6 | Fig. 3 and 4 in Ref. [67] | 100 mM $H_2SO_4$<br>200 mM KOH | 0.22 | + → –, o | + → –, o |
| 7 | Fig. 1A, 1B & 7 in Ref. [68] | 10 mM Glycine<br>10 mM Arginine | –0.27 | + ← –, o | + ← –, o |
| 8 | Fig. 2 in Ref. [68] | 10 mM Glycine<br>10 mM Lysine | 1.91 | + → –, o | + → –, o |
| 9 | Fig. 1, 2 and 3 in Ref. [59] | 4.25% Phosphoric acid<br>5.0% Ethylenediamine | 0.02 | + → –, w | + → –, w |
| 10 | Fig. 1 in Ref. [63] | 10 mM Aspartic acid<br>10 mM Lysine | 2.35 | + → –, o | + → –, o |
| 11 | Fig. 2, 3A, 3C in Ref. [64] | 10 mM MES<br>10 mM GABA | $1.58\times10^{8}$ | + → –, s | + → –,s |
| 12 | Fig. 3B in Ref. [64] | 750 mM MES<br>750 mM GABA | $1.37\times10^{10}$ | + → –, s | + → –, s |
| 13 | Fig. 1A, 2A in Ref. [65] | 10 mM Aspartic acid<br>10 mM Histidine | 116 | + → –, s | + → –, s |
| 14 | Fig 5 in Ref. [65] | 10 mM Acetic acid<br>10 mM Tris | 0.02 | + → –, w | + → –, w |
| 15 | Fig. 6 in Ref. [66] | 40 mM Glutamic acid<br>40 mM Histidine | 270 | + → –, s | + → –, s |
| 16 | Fig. 3 in Ref. [66] | 10 mM Threonine<br>10 mM Lysine | 2.36 | + → –, o | + → –, o |
| 17 | Fig. 1 in Ref. [66] | 10 mM Threonine<br>10 mM Histidine | 116 | + → –, s | + → –, s |
| 18 | Fig. 2 in Ref. [66] | 10 mM Glycine<br>10 mM Histidine | 100 | + → –, s | + → –, s |
| 19 | Fig. 4 in Ref. [66] | 10 mM Glutamic acid<br>10 mM Histidine | 121 | + → –, s | + → –, s |

| | | | | | |
|---|---|---|---|---|---|
| 20 | Fig. 2 in Ref. [71] | 100 mM acetic acid<br>100 mM Tris | 0.01 | + → –, w | + → –, w |
| 21 | Fig. 1A, 1B in Ref. [58] | 0.02% $H_2SO_4$<br>0.40% Ethylenediamine | –0.02 | + ← –, w | + → –, w |

[a)] The computation of Rr value is the same as those in Table 3.

[b)] "+ ← –" means the anodic drifting of pH gradient; "+ → –" implies the cathodic drifting of pH gradient; w = weak; o = obvious; s = strong.

**Table S2**. Agreements between the predictions of $R_r$ values and the experiments of Hjerten's mobilization of pH gradient induced by adding salt after IEF in references.[a)]

| No. | IEF run | Acid_base pairs | $R_r$ | Prediction | Experiment |
|---|---|---|---|---|---|
| 1 | Table 1 in Ref. [80] | 20 mM NaOH<br>20 mM NaOH | $-1.583\times10^{10}$ | Anodic | Anodic |
| 2 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$+80 mM NaCl<br>20 mM NaOH | –2.110 | Anodic | Anodic |
| 3 | Table 1 in Ref. [80] | 20 mM pH 3.6 $Na_3PO_4$<br>20 mM NaOH | –34.36 | Anodic | Anodic |
| 4 | Table 1 in Ref. [80] | 20 mM pH 6.8 $Na_3PO_4$<br>20 mM NaOH | $-6.03\times10^{4}$ | Anodic | Anodic |
| 5 | Table 1 in Ref. [80] | 20 mM pH 11.5 $Na_3PO_4$<br>20 mM NaOH | $-3.55\times10^{9}$ | Anodic | Anodic |
| 6 | Table 1 in Ref. [80] | 20 mM pH 7.0 EA[b)] –$H_3PO_4$<br>20 mM NaOH | $-3.17\times10^{4}$ | Anodic | Anodic |
| 7 | Table 1 in Ref. [80] | 20 mM pH 11 EA[a)]<br>20 mM NaOH | $-4.92\times10^{7}$ | Anodic | Anodic |
| 8 | Table 1 in Ref. [80] | 20 mM pH 6.8 $Na_3PO_4$<br>20 mM pH 9.0 Gly–NaOH | $-6.77\times10^{2}$ | Anodic | Anodic |
| 9 | Table 1 in Ref. [80] | 20 mM NaCl<br>20 mM pH 9.0 Gly–NaOH | $-5.22\times10^{2}$ | Anodic | Anodic |
| 10 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>20 mM $H_3PO_4$ | $1.33\times10^{10}$ | Cathodic | Cathodic |
| 11 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>100 mM NaCl | $5.66\times10^{5}$ | Cathodic | Cathodic |
| 12 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>20 mM pH 6.8 $Na_3PO_4$ | $3.52\times10^{5}$ | Cathodic | Cathodic |
| 13 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>20 mM NaOH+20 mM NaCl | 0.932 | Cathodic | Cathodic |
| 14 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>20 mM NaOH+40 mM NaCl | 1.702 | Cathodic | Cathodic |
| 15 | Table 1 in Ref. [80] | 20 mM $H_3PO_4$<br>20 mM NaOH+80 mM NaCl | 3.373 | Cathodic | Cathodic |
| 16 | Fig. 2a, 3 and 4 in Ref. [78] | 20 mM NaOH<br>20 mM NaOH | $-1.583\times10^{10}$ | Anodic | Anodic |
| 17 | Fig. 2b in Ref. [78] | 20 mM $H_3PO_4$<br>20 mM $H_3PO_4$ | $1.33\times10^{10}$ | Cathodic | Cathodic |
| 18 | Fig. 1, 3 and 5 in Ref. [82] | 20 mM NaOH<br>20 mM NaOH | $-1.58\times10^{10}$ | Anodic | Anodic |

| | | | | | |
|---|---|---|---|---|---|
| 19 | Fig. 4 in Ref. [82] | 20 mM $H_3PO_4$<br>20 mM $H_3PO_4$ | $1.33\times10^{10}$ | Cathodic | Cathodic |
| 20 | Fig. 9 in Ref. [81] | 20 mM $H_3PO_4$<br>10 mM Glysine | 123.7 | Cathodic | Cathodic |
| 21 | Fig.1,2,4B,5B ,6B in ref. 87 | 10 mM $H_3PO_4$<br>10 mM acetic acid | $2.92\times10^{7}$ | Cathodic | Cathodic |
| 22 | Fig. 1A in Ref. [88] | 200 mM acetic acid<br>2000 mM ammonium acetate | $1.31\times10^{7}$ | Cathodic | Cathodic |
| 23 | Fig.2,4 in Ref. [89] | 100 mM $H_3PO_4$<br>40 mM NaOH + 80 mM NaCl | 1.81 | Cathodic | Cathodic |
| 24 | Fig. 1 – 5 in Ref. [92] | 91 mM $H_3PO_4$<br>20 mM pH 11.85 NaOH–$H_3PO_4$ | 0.841 | Cathodic | Cathodic |
| 25 | Fig. 5,11,12, 14 in ref. [90] | 100 mM $H_3PO_4$<br>20 mM NaOH + 20 mM NaCl | 0.844 | Cathodic | Cathodic |
| 26 | Fig. 9 in ref. [91] | 10 mM $H_3PO_4$<br>20 mM NaOH+5 mM formic acid | 0.407 | Cathodic | Cathodic |
| 27 | Fig. 9 in Ref. [91] | 10 mM $H_3PO_4$<br>20 mM NaOH + 5 mM $H_3PO_4$ | 0.912 | Cathodic | Cathodic |
| 28 | Fig. 9 in Ref. [91] | 10 mM $H_3PO_4$<br>20 mM NaOH + 5 mM HCl | 0.449 | Cathodic | Cathodic |
| 29 | Fig. 10 in Ref. [91] | 10 mM $H_3PO_4$<br>20mM NaOH+10mM formic acid | 0.746 | Cathodic | Cathodic |
| 30 | Fig. 10 in ref. [91] | 10 mM $H_3PO_4$<br>20 mM NaOH+10 mM $H_3PO_4$ | 343.6 | Cathodic | Cathodic |
| 31 | Fig. 10 in Ref. [91] | 10 mM $H_3PO_4$<br>20 mM NaOH+10 mM HCl | 0.866 | Cathodic | Cathodic |
| 32 | Ref. [92] | 100 mM $H_3PO_4$<br>40 mM NaOH | 0.218 | Cathodic | Cathodic[c)] |
| 33 | Ref. [93] | 10 mM $H_3PO_4$<br>20 mM NaOH | 0.237 | Cathodic | Cathodic[c)] |

[a)] The computation of $R_r$ value is the same as that in Table 3.

[b)] EA = Ethanolamine.

[c)] Results of computer simulations.