A Novel and Optimized Method for Electro-focusing and Moving Neutralization Reaction Boundary Formed by HCl and NaOH

Shu Lin ZHOU^{1,2}, Cheng Xi CAO¹*, You Zhao HE¹, Wen Kui CHEN², Yi Tai QIAN¹

¹Department of Chemistry, University of Science and Technology of China, Hefei 230026 ²Institute of Allergy Research, Wannan Medical College, Anhui Wuhu 241001

Abstract: A novel method is developed for electro-focusing and moving neutralization reaction boundary (MNRB) created with HCl and NaOH. The optimized conditions are screened out. By using this method, the experiments are performed on MNRB formed with HCl and NaOH in agarose gel. The experiments are quantitatively in coincidence with the predictions with the theory of moving chemical reaction boundary (MCRB).

Keywords: Electrolyte, Electrolysis, Electromigration, Isoelectric focusing (IEF), Moving chemical reaction boundary (MCRB).

The theory of MCRB has been developed by C. X. $\text{Cao}^{1.4}$, Deman-Rigole^{5,6} and Pospichal *et al.*⁷. The relations between the MCRB and IEF^{8,9} have also been revealed by C. X. Cao *et al.*¹⁰⁻¹³. The theory has been partially demonstrated by the experiments performed by the above workers^{5-7,14-16}.

But, the validity of the theory for MNRB has not been manifested, and efficient method has not been developed for the MNRB and electro-focusing that is based on the mechanism of MNRB¹⁰⁻¹². The purposes of this work are to supply a novel method, screen out the optimized experimental conditions and test the validity of the theory.

Experimental

The apparatus is homemade as shown in **Figure 1**. A glass tube is filled agarose gel containing NaOH, 0.1 mol/L KCl and 0.1% (w/v) bromophenol blue. The tube, coupled with the ruler, is fixed on a small performing-table, over which an adjustable camera (Model SD345, Minolta Co. Ltd., Japan) is fixed. The camera is used to record the movement of MNRB with the time. With the two rubber-tubes, the two ends of the glass tube are connected to two three-way-pipes, which are joined with two peristaltic pumps (*viz.*, pump 1 and 2, model HL-2, Shanghai Huxi Biochemical Instrument Factory, Shanghai) and two platinum electrodes (*viz.*, the anode and cathode). A power supply (model DYY-III4, Beijing Luyi Instrument Factory, Beijing) is used to yield the direct current. At the two ends of glass tube, two electrodes inserted into the rubber tubes are connected with a recorder (XWD2-213, Shanghai Automatic Instrument Factory No. 3,

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Shanghai), with which one monitors the voltage between the two ends of the glass tube.

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Figure 1 The apparatus and procedure for the experiments of MNRB

The arrows mean the flow directions of anolyte and catholyte, the symbols, + and -, mean the anode and cathode respectively.

Initially, the agarose gel contains 0.004-0.014 mol/L NaOH, 0.1 mol/L KCl and 0.1% bromophenol blue. The catholyte holds 0.004-0.014 mol/L NaOH and 0.1 mol/L KCl. The anolyte comprises 0.004-0.014 mol/L HCl and 0.1 mol/L KCl. All MNRBs move towards the cathode. Thus, before run, the bromophenol in the overall tube is blue due to the existence of NaOH (**Figure 2A**). However, after the run the left end of the tube turns yellow (**Figure 2B**), when the HCl displaces the NaOH gradually. As the displacement goes, the yellow zone becomes longer, conversely the blue zone is shorter. A clear boundary is observed during the electromigration reaction between H⁺ and OH. Here, the bromophenol is a mark for the yellow acidic and blue alkaline zones and the boundary that separates the yellow and blue zones sharply (**Figure 2**). The photographs are taken with the time (*t*) during the run, as shown in **Figure 2**. With the photographs, one can directly monitor the displacements, *viz.*, the length (l_{obs}) of yellow zone. The theoretic and observed velocities are computed with Eqn. (1) and (2), respectively¹⁴.

$$\mu_{\text{theo}}^{\alpha\beta} = \frac{m_{\text{act, H}+}^{\alpha} c_{\text{H}+}^{\alpha} - m_{\text{act, OH}-}^{\beta} c_{\text{OH}-}^{\beta}}{\left(c_{\text{H}+}^{\alpha} - c_{\text{OH}-}^{\beta}\right)\kappa} i$$
(1)

$$\mu_{\rm obs}^{\alpha\beta} = l_{\rm obs} \,/\, t \tag{2}$$

where, *i* is the current intensity, κ the specific conductivity, μ the boundary velocity, *m* the mobility and *c* the concentration in equiv./L. The superscripts, α , β and $\alpha\beta$ mean phase α and β , and boundary $\alpha\beta$ respectively, the subscripts, theo and obs, reply the theoretic and observed velocities respectively, the subscripts, H+ and OH-, indicate the H⁺ and OH respectively, and subscript, act, means the actual mobility.

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Figure 2 The MNRB created with HCl and NaOH A: the initial state just before run B: 5 minutes after the run C: 10 minutes after the run.



The arrows indicate the boundary; the symbols of + and - mean the anode and cathode respectively; the yellow zone ranges from the symbol + to the arrows; the blue zone from the arrows to the symbol -. Conditions: 0.01 mol/L HCl, 0.008 mol/L NaOH, 0.1 mol/L KCl, 1% (w/v) agarose gel, 0.1% bromophenol blue, flow rates 0.6 ml/min, constant current intensity 0.6 mA/mm², I.D. 4.5 mm and length 90 mm, temperature 18-20°C.

Results

We have investigated several influence factors and optimized the conditions as follow: (1) current intensity(CI) 0.4-0.8 mA/mm², (2) product of time and CI less than 54-60 $s \cdot mA/mm^2$, (3) temperature 18-20°C, (4) agarose gel 1%(w/v), (5) fresh solution and gel containing NaOH, and (6) photographing rightly over the boundary.

Totally, 18 runs were performed under the above optimized conditions. The statistic analysis of the 18 runs is given in **Figure 3**. It is clearly shown that the correlation coefficient (CC) between the observed and theoretic velocities is equal to 0.9988 (n=18) and the regression equation is Y = 0.9912X - 0.0058 (Y= the theoretic velocity and X = the observed). The experiments are in good agreement with the theory of MCRB. Thus, the results proved the validity of the theory for a MNRB created by HCl and NaOH.





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All of the runs are performed under the circumstance temperature of 18-20°C.

Acknowledgments

The project is supported by the NSFC (No. 29775014) and Chinese Health Committee (98-2-334).

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Received 14 March, 2000 Revised 31 October, 2000