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The invalidity of Kohlrausch' regulating function for Svensson's isoelectric focusing and stationary electrolysis at steady state

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Abstract

Kohlrausch' regulating function is of important significance in the field of electrophoresis. In this paper, the relative regulating function is defined from Kohlrausch' regulating function. The relative values, including the limited values, of the regulating function for the stationary electrolysis of salt, on which the classic isoelectric focusing (IEF) is based, are computed and compared with the computer program of the QBASIC written by us. The results directly demonstrate that, (1) in a few cases the regulating function is valid for the stationary electrolysis and IEF, whereas (2) the function is, in most of cases, not valid for the stationary electrolysis and IEF at steady-state. Those findings may be useful for the studies on the relationships between Kohlrausch' regulating function and IEF and for the classification of numerous electrophoretic techniques. © 1999 Published by Elsevier Science B.V. All rights reserved.

Keywords: Kohlrausch' regulating function; Moving boundary system; Isoelectric focusing; Isotachopheresis

1. Introduction

A universal important function in the field of electrophoresis is the Kohlrausch' regulating function [1] (see Eq. (2) here). The regulating function is clearly valid for zone electrophoresis (ZE) due to the existence of homogeneity of electrolyte solution in ZE.

The function is also valid for moving boundary electrophoresis and isotachopheresis (ITP). With the

regulating function, Kendall and Crittenden [2,3] invented the ionic migration method, viz., the early ITP, to separate rare-earth metal ions and some simple acids. With this function, MacInnes and Longworth [4] developed the moving boundary method (MBM) to detect the transference numbers of small ions, an important physico-chemical parameter with which, joined with equivalent conductivity of electrolyte solution, one can compute ionic mobility [5–8]. With the function, Everaerts and Martin [9] studied the moving boundary electrophoresis, which lead to the birth of iso-tacho-electrophoresis (the Greek iso=equal, tacho=velocity), viz., iso-tacho-phoresis (ITP), an important electrophoretic technique.

The regulating function is also used for studies on

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isoelectric focusing (IEF). Bier, Mosher and Thormann [10–14] used the regulating function to perform a computer simulation of IEF, to reveal the experimental and theoretical dynamics of IEF and to elucidate the dynamic mechanism of anodic and cathodic drifts of pH gradient in IEF. Pospichol, Deml and Bocek [15,16] also used the regulating function to perform a computer simulation of IEF and electrically controlled electro-focusing. It was found that the computer simulations based upon the function were quantitatively in agreement with the experiments of electrically controlled electro-focusing. Those studies show the extent of validity of the function for IEF.

However, the authors find that the function is not valid for some special cases, such as the stationary electrolysis of salt on which the classic IEF is based. Here, we report the findings.

2. Notations and definitions

c : the equivalent concentration (equiv./m³). The subscripts, + and –, indicate the positive and negative reaction ions respectively and the superscripts α and β imply phase α and β respectively. Signed quantity, positive if the ion carries net positive charge(s), and negative if net negative charge(s).

m : the mobility (m² s⁻¹ V⁻¹). The subscripts + and – indicate the positive and negative reaction ions respectively, the subscripts, 0 and act, imply the absolute and actual ionic mobilities respectively, and the superscripts α and β imply phase α and β respectively. Signed quantity.

F : the Faraday constant (=96 500 coulombs per equivalent).

ω : the symbol of Kohlrausch' regulating function (mol V s m⁻⁵), viz., the omega function. The subscript, rel, indicates the relative regulating function.

z : ionic charge number (dimensionless).

λ_0 : the limited molar conductivity at 25°C (S⁻² m² mol⁻¹)

$$I: \text{ionic strength (mol/l)} = 0.5 \sum c_i z_i^2 \quad (1)$$

η : the coefficient. If $z=1$, $\eta=0.5$, and if $z=2$, or 3, or 4, or ..., $\eta=0.77$.

3. Brief theory

The regulating function, as formulated by Kohlrausch [1], is given as

$$\omega = \sum_i \frac{c_i^\alpha}{m_i^\alpha} = \sum_i \frac{c_i^\beta}{m_i^\beta} \quad (2)$$

The function shows that, for a steady-state boundary, the sums of the ionic concentrations divided by their mobilities on either side homogeneity solutions of a boundary are the same values and instead constant as a function of time and location. In other words, the values of the function on two sides of a boundary are equal to each other at any time and at any place.

In order to compare the values of the regulating function in different solutions/or phases, we should define the relative regulating function from Eq. (2).

If $\omega^\alpha > \omega^\beta$, we define the relative regulating function as

$$\omega_{\text{rel}} = 1 - \frac{\omega^\beta}{\omega^\alpha} = 1 - \frac{\sum_i c_i^\beta / m_i^\beta}{\sum_i c_i^\alpha / m_i^\alpha} \quad (3a)$$

whereas, if $\omega^\alpha < \omega^\beta$, we define

$$\omega_{\text{rel}} = \frac{\omega^\alpha}{\omega^\beta} - 1 = \frac{\sum_i c_i^\alpha / m_i^\alpha}{\sum_i c_i^\beta / m_i^\beta} - 1 \quad (3b)$$

Eqs. (3a) and (3b) are the relative comparisons between the values of Kohlrausch' regulating function in two phases/or solutions in an electrophoretic system. Thus, they should be called as the relative regulating function. Evidently, the significance of $\omega_{\text{rel}} \cong 0$ (such as -0.05, 0 and 0.05 as given in Table 1) is that the values of the regulating function in one phase is equal/or very close to that in another phase. In other words, the regulating function does hold in a boundary system. That of $\omega_{\text{rel}} \neq 0$ implies that the values in the two phases are not equal to each other. Namely, the regulating function does not persist for a boundary system. For more detailed information, see Table 1.

4. Literature data and treatments

As have been pointed out by Svensson [17,18] and

Table 1

The significance of different relative values of Kohlrausch' regulating function (ω_{rel})

$\omega_{\text{rel}} =$	$\rightarrow 1$	0.95	0.90	0.75	0.50	0.25	0.10	0.05	0
$\omega^\alpha/\omega^\beta =$	$\rightarrow \infty$	20.00	10.00	4.00	2.00	1.33	1.11	1.05	1.00
$\omega^\alpha/\omega^\beta =$	$\rightarrow 0$	0.05	0.10	0.25	0.50	0.75	0.90	0.95	1.00

being continuous

$\omega_{\text{rel}} =$	-0.05	-0.10	-0.25	-0.50	-0.75	-0.90	-0.95	$\rightarrow -1$
$\omega^\alpha/\omega^\beta =$	0.95	0.90	0.75	0.50	0.25	0.10	0.05	$\rightarrow 0$
$\omega^\beta/\omega^\alpha =$	1.05	1.11	1.33	2.00	4.00	10.00	20.00	$\rightarrow \infty$

Tiselius [19], during stationary electrolysis of salt, the acid is accumulated in the anodic vessel, whereas the alkali is collected in the cathodic vessel. The classic IEF, viz., Svensson's IEF, is based on the stationary electrolysis of salt, which can simply be achieved with an acid and a base as the analyte and catholyte, respectively. As studied by the authors [20–23], a neutralization reaction boundary is created during the stationary electrolysis. The boundary is stationary at steady-state. Therefore, the stationary electrolysis, on which the classic IEF is based and which can be achieved with an acid and alkali as the anolyte and catholyte respectively, can be treated with Eqs. (3a) and (3b).

The systems of stationary electrolysis, considered here, are HCl–KOH, HCl–NaOH, H₂SO₄–KOH and H₂SO₄–NaOH. It is not necessary to perform some experiments to obtain the basic data, because the basic data can be obtained from the literature data [24]. In Table 2, the absolute ionic mobilities in

infinitely diluted solutions is obtained from limited ionic conductivities [24] with

$$m_0 = \frac{\lambda_0}{F} \quad (4)$$

The actual ionic mobilities in different solutions with different concentrations are figured from the absolute ionic mobilities with the empirical equation

$$m_{\text{act}} = m_0 \exp(-\eta\sqrt{zI}) \quad (5)$$

$(z = 1, \eta = 0.5; z = 2, 3, 4, \dots, \eta = 0.77)$

which is valid for both large ions with low electrical charge intensities [5,6] and small ions, including hydrogen ion, with very high electrical charge intensities [7,8], if the ionic strength is less than 0.1 mol/l.

With the data in Table 2, we can easily calculate the values of Kohlrausch' regulating function for the

Table 2

The absolute and actual ionic mobilities ($\times 10^{-8} \text{ m}^2 \text{ s}^{-1} \text{ V}^{-1}$) at different concentrations at 25°C

Concentration (mol/l)	infinite ^a	0.0001 ^b	0.0005 ^b	0.001 ^b	0.005 ^b	0.01 ^b	0.05 ^b
H ⁺ (in HCl)	36.23	36.05	35.83	35.66	34.97	34.46	32.40
Cl ⁻ (in HCl)	7.91	7.87	7.82	7.79	7.63	7.54	7.07
K ⁺ (in KOH)	7.62	7.58	7.54	7.50	7.36	7.25	6.81
Na ⁺ (in NaOH)	5.19	5.16	5.13	5.11	5.01	4.94	4.64
OH ⁻ (in KOH/NaOH)	20.52	20.42	20.29	20.20	19.81	19.52	18.35
H ⁺ (in H ₂ SO ₄)	36.23	35.92	35.54	35.25	34.08	33.22	29.85
SO ₄ ²⁻ (in H ₂ SO ₄)	8.29	8.18	8.05	7.95	7.54	7.25	6.15

^a The absolute ionic mobilities are calculated from the limited molar conductivities of H⁺ (349.65), K⁺ (73.48), Na⁺ (50.08), OH⁻ (198.0), SO₄²⁻ (80.0) and Cl⁻ ($76.31 \times 10^{-2} \text{ S m}^2 \text{ mol}^{-1}$ at 25°C) [24] with Eq. (4).

^b The actual ionic mobilities are computed from the absolute mobilities with Eq. (5).

electrolyte systems of HCl–KOH, HCl–NaOH, H₂SO₄–KOH and H₂SO₄–NaOH discussed below.

It should be noticed here, in the electrolyte systems formed with an acid and alkali, the solution of acid like HCl/H₂SO₄ discussed here is treated as phase α , and that of base, for instance NaOH/KOH, as phase β . All of the calculations are performed with the computer program written by the authors with the QBASIC (Ver. 4.5, Microsoft, Redmond, WA, USA) running on a personal computer (486DX-80, 16 M byte RAM, 540 M byte hard disk, Copam⁺ Electronics, Taiwan, China). The calculated data are all compared with those computed with the software of EXCEL(ver. 5.0, Microsoft, Redmond, WA, USA), no differences are found between the former and the latter.

5. Results

5.1. Limited values of Kohlrausch' regulating function

In this paper, we consider those electrolyte systems of HCl–KOH, HCl–NaOH, H₂SO₄–KOH and H₂SO₄–NaOH, which can be achieved by the stationary electrolysis of salts of KCl, NaCl, K₂SO₄ and Na₂SO₄ respectively. First, we compute the limited values of the relative regulating function for those electrolyte systems diluted infinitely with the following limited regulating function

$$\lim_{c_i^\alpha, c_i^\beta \rightarrow 0} (\omega_{\text{rel}}) = \omega_{\text{rel},0} = 1 - \frac{\omega_0^\beta}{\omega_0^\alpha}$$

$$= 1 - \frac{\sum_i c_{i,0}^\beta / m_{i,0}^\beta}{\sum_i c_{i,0}^\alpha / m_{i,0}^\alpha} \quad (\text{for } \omega^\alpha > \omega^\beta) \quad (6a)$$

$$\lim_{c_i^\alpha, c_i^\beta \rightarrow 0} (\omega_{\text{rel}}) = \omega_{\text{rel},0} = \frac{\omega_0^\alpha}{\omega_0^\beta} - 1$$

$$= \frac{\sum_i c_{i,0}^\alpha / m_{i,0}^\alpha}{\sum_i c_{i,0}^\beta / m_{i,0}^\beta} - 1 \quad (\text{for } \omega^\alpha < \omega^\beta) \quad (6b)$$

With Eq. (6), the limited values of regulating function are estimated for the electrolyte systems, A=HCl–KOH, B=HCl–NaOH, C=H₂SO₄–KOH and D=H₂SO₄–NaOH, respectively. Those limited values are –0.1442, –0.3620, –0.0228 and 0.2717 as given in Fig. 1. Obviously, the limited values for A, B and D are not close to zero, except for C (= –0.0228) which is very near to zero.

5.2. Relative values for the equal-concentration electrolyte systems

By using Eq. (5), we can compute the relative values of regulating function for those electrolyte systems of HCl–KOH, HCl–NaOH, H₂SO₄–KOH

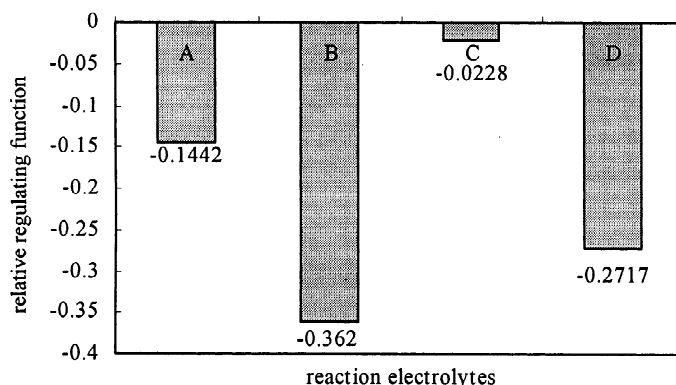


Fig. 1. The limited values of relative Kohlrausch' regulating function for those reaction electrolytes of A=HCl–KOH, B=HCl–NaOH, C=H₂SO₄–KOH and D=H₂SO₄–NaOH.

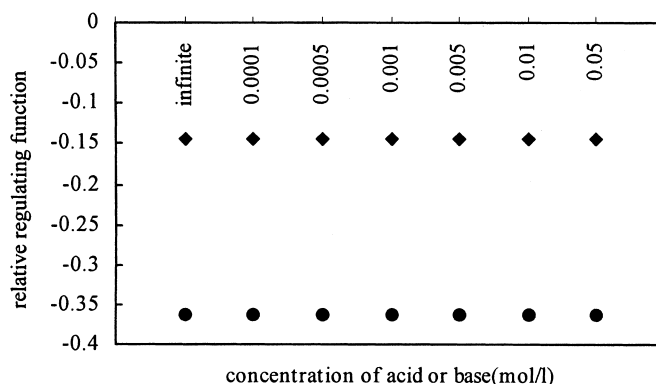


Fig. 2. The relative values of Kohlrausch' regulating function for the equal-concentration electrolyte systems of KCl-KOH (◆) and HCl-NaOH (●).

and H_2SO_4 -NaOH with the equal-concentration acid HCl/or H_2SO_4 and base KOH/or NaOH.

Fig. 2 shows the results of relative values for the electrolyte systems of KCl-KOH (◆) and HCl-NaOH (●). It was clearly displayed by Fig. 2 that the values for HCl-KOH and HCl-NaOH are respectively -0.1442 and -0.3619 , which are constant with the synchronous enormous increases of HCl and NaOH/KOH from the infinitely diluted solution to the 0.05 mol/l.

As given in Fig. 2, the relative regulating function values are shown to be well below zero. This means the values of Kohlrausch' regulating function in HCl solution are certainly different from that in KOH/or NaOH solution, even the concentrations of HCl and KOH/or NaOH are equal to each other. The con-

dition of equal-concentration always occurs in an IEF, because during the run of IEF an equal-concentration of acid and alkali are used as the catholyte and anolyte respectively [17–23].

Fig. 3 manifests the relative values of Kohlrausch' regulating function for the equal-concentration electrolyte systems of H_2SO_4 -KOH (◆) and H_2SO_4 -NaOH (●). It is proved in Fig. 3 that, (1) the relative values for the equal-concentration of H_2SO_4 -KOH (◆) is in the range from -0.0228 to 0.1232 , very near to zero especially for the concentration range from zero mol/l to 0.01 mol/l, this results is evidently different from those in Fig. 2 for the equal-concentration of HCl-KOH; (2) whereas, the relative values for H_2SO_4 -NaOH (●) are not close to zero (very similar to those in Fig. 2 for HCl-

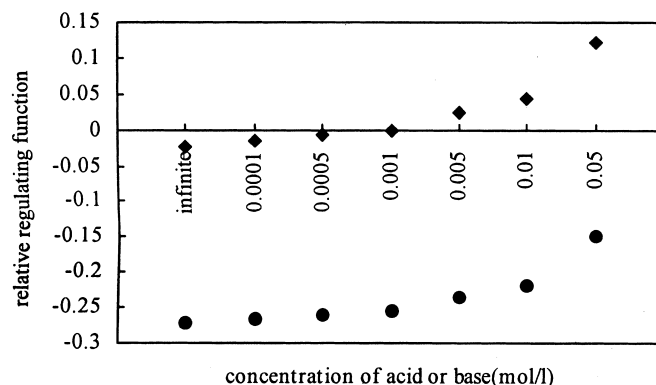


Fig. 3. The relative values of Kohlrausch' regulating function for the equal-concentration electrolyte systems of H_2SO_4 -KOH (◆) and H_2SO_4 -NaOH (●).

NaOH); and (3) the relative values for both H_2SO_4 –KOH and H_2SO_4 –NaOH are slowly increased as the concentrations of the acid H_2SO_4 and base KOH/NaOH increase synchronously, the results are quite different from those in Fig. 2 due to the existence of constant relative values.

Evidently, under the conditions with the equal-concentration acid and base as phase α and β , respectively, there exists the situation where $\omega_{\text{rel}} \approx 0$, which means the values of the regulating function in two phases are almost equal to each other, while there is, in most of cases, the $\omega_{\text{rel}} \neq 0$ which implies the un-validity of the regulating function.

5.3. Relative values for the different-concentration electrolyte systems

Lastly, we calculate the relative values of Kohlrausch' regulating function for the above electrolyte systems with different molar concentrations. Fig. 4 is for the electrolyte systems of HCl–NaOH, the concentrations of HCl and NaOH change from 0.0001 to 0.05 mol/l. Figs. 5–7 are for HCl–KOH, H_2SO_4 –NaOH and H_2SO_4 –KOH respectively, their concentration ranges are the same as those of Fig. 4.

As is shown in Fig. 4, if the concentration of NaOH is fixed at a value, for example 0.001 mol/l, the relative value of regulating function increases almost from -1 to 1 with the increase of the HCl concentration from 0.0001 to 0.05 mol/l, and if

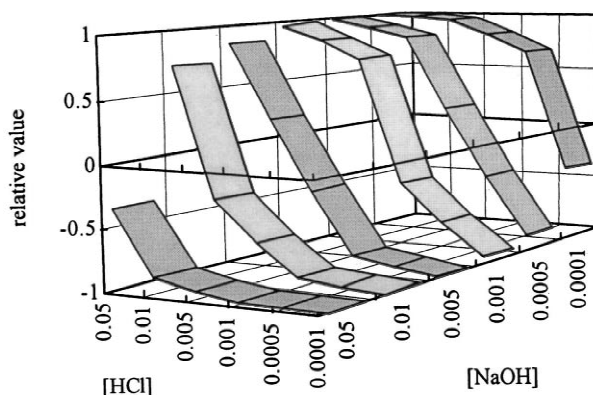


Fig. 4. The relative values of Kohlrausch' regulating function for the different concentration electrolyte system of HCl–NaOH. [NaOH] and [HCl] mean the molar concentrations of NaOH and HCl respectively.

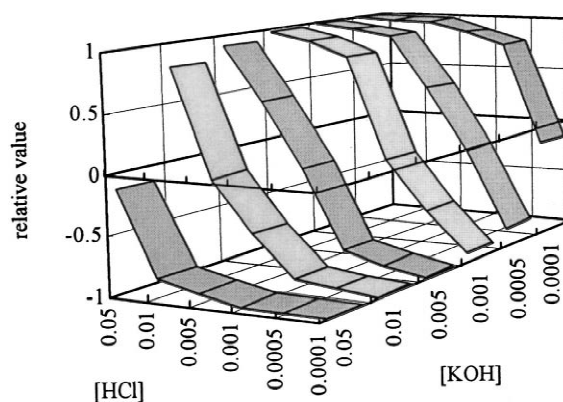


Fig. 5. The relative values of Kohlrausch' regulating function for the different-concentration electrolyte system of HCl–KOH. [KOH] and [HCl] mean the molar concentrations of KOH and HCl respectively.

[HCl] is constant, whereas, the relative value decreases with the increase of the concentration of NaOH. Obviously, one can find most of the curves, except for that of $[\text{NaOH}] = 0.05$ mol/l, are across the “zero square” of relative value, which means that the relative value may be equal/or very close to zero (or between -0.05 and 0.05) if similar concentrations of NaOH and/or HCl are selected. However, most of the values are clearly away from the “zero square”.

Those observations are also present in those electrolyte systems of HCl–KOH (see Fig. 5),

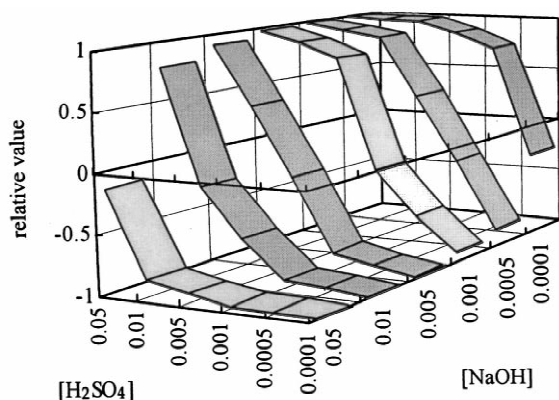


Fig. 6. The relative values of Kohlrausch' regulating function for the different-concentration electrolyte system of H_2SO_4 -NaOH. $[\text{NaOH}]$ and $[\text{H}_2\text{SO}_4]$ indicate the molar concentrations of NaOH and H_2SO_4 respectively.

H_2SO_4 -NaOH (see Fig. 6) and H_2SO_4 -KOH (Fig. 7).

As compared with the systems of HCl-NaOH, HCl-KOH and H_2SO_4 -NaOH, there is a difference in H_2SO_4 -KOH shown in Fig. 7. The difference is all curves are across the "zero square" of relative value.

To conclude, there are two results. The first result is the $\omega_{\text{rel}} \approx 0$, viz., near/or equal to the "zero square", as shown in Figs. 4–7, this result implies the validity of the regulating function for those

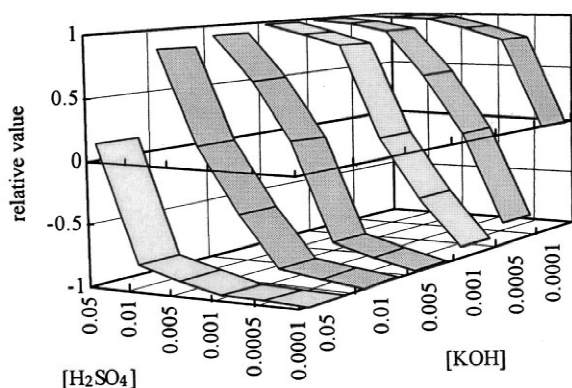


Fig. 7. The relative values of Kohlrausch' regulating function for the different-concentration electrolyte system of H_2SO_4 -KOH. $[\text{KOH}]$ and $[\text{H}_2\text{SO}_4]$ the molar concentrations of KOH and H_2SO_4 respectively.

electrolyte systems. However, the second, as is observed in Figs. 4–7, is the $\omega_{\text{rel}} \neq 0$, which indicates the non-validity of the regulating function for the stationary electrolysis of some salts and IEF at steady-state.

6. Discussions and conclusions

Stationary neutralization boundary is present in stationary electrolysis of salt and IEF at steady state. As stated by Svensson and Tiselius [17–19], during the stationary electrolysis of sodium sulfate, NaOH is collected in the anodic vessel, while sulfuric acid is accumulated in the cathodic cell, the classic IEF is based on the stationary electrolysis which may be simply achieved by using an acid and a base as the catholyte and anolyte, respectively. Under the electric field, the hydrogen ion in the catholyte moves towards the anode, but the hydroxyl ion in the anolyte migrates towards the cathode. Obviously, a neutralization boundary is formed in stationary electrolysis due to the electromigration reaction between hydrogen and hydroxyl ions migrating in opposite directions. The boundary is, as pointed out by Svensson [17–19], stationary due to stationary electrolysis, this was further proved by the authors [21–23] with the existence of quasi/equal transference numbers of hydrogen and hydroxyl ions in the stationary electrolysis and classic IEF.

Kohlrausch' regulating function is not valid for the stationary electrolysis of some salts and IEF at steady-state. With the analytical data of the stationary electrolysis and IEF obtained with the relative Kohlrausch' regulating function as given in Sections 4 and 5, the paper verifies that, in a few cases the regulating function is valid for IEF [17–23] due to the existence of $\omega_{\text{rel}} = / \text{or} \approx 0$, viz., the "zero square", as shown in Figs. 2–7. However in most cases the regulating function is not valid for the stationary electrolysis, on which the classic IEF is based. To the authors' knowledge, the results have not been reported. The findings may be useful for the studies on the relationships between Kohlrausch' regulating function and IEF and for studies on the classification of numerous electrophoretic techniques.

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