

CHROM 21 215

## METHODS FOR ON-LINE DETERMINATION AND CONTROL OF ELECTROENDOSMOSIS IN CAPILLARY ELECTROCHROMATOGRAPHY AND ELECTROPHORESIS<sup>a</sup>

B. J. WANDERS,\* A. A. M. VAN DE GOOR and F. M. EVERAERTS

*Laboratory of Instrumental Analysis, Eindhoven University of Technology, P. O. Box 513, 5600 MB Eindhoven (The Netherlands)*

---

### SUMMARY

Systems are described for the on-line determination and control of electroendosmosis that offer several advantages for both electrochromatographic and electrophoretic separations. For electrochromatographic separations the control system can be used to stabilize the electroendosmosis and obtain a better reproducibility of the results. For electrophoretic separations this control system offers the possibility of a so-called "upstream" separation, which results in a decrease in the plate height and thus a better resolution. Further, the controllable electroendosmosis can be used to determine with high reproducibility, in one run, both ionic (anions and cations) and non-ionic components, using only one detector.

---

### INTRODUCTION

Electroendosmotic flow (EOF) is a liquid flow that occurs if an electric field is applied over the length of a capillary that has an active surface and is filled with an electrolyte. This flow originates from the existence of an electric double layer between the charged capillary wall and the liquid. The theoretical background has been discussed more extensively elsewhere<sup>1-7</sup>.

In capillary zone-electrophoresis (CZE), in which ionic components are separated on basis of the difference in their effective mobilities in an electric field, and in capillary electro-chromatography (CEC, also called electroendosmotically driven liquid chromatography<sup>8-10</sup>), in which non-ionic components are separated, EOF can be used as a liquid pump. Instead of open capillaries, capillaries packed with small particles or tubes can also be applied<sup>11</sup>.

In all these instances it is of the greatest importance that the EOF is constant, or at least is known during the experiment (for the identification of the components). A solution to this problem is an on-line measurement and control system for EOF, as described here.

---

<sup>a</sup> Patent pending, application No. 8802273 (The Netherlands).

## THE CONTROL SYSTEM

The control system for electroendosmosis consists of two main parts. The first part is a unit which measures the EOF on-line and the value is registered by a computer. The second part is the control unit, which can execute control both directly and indirectly.

*The measurement unit*

*On-line determination of EOF using an analytical balance.* With the use of an analytical micro-balance, the weight of one electrode vessel is monitored in a specified time interval (1–10 s) and this value is sent to a computer through an interface (IEEE or RS232) (Fig. 1). With this value the EOF can be calculated using the equation.

$$v_{eo} = \Delta W / \Delta t \cdot 1 / (\pi r^2 \rho) \quad (1)$$

where  $v_{eo}$  is the EOF (m/s),  $\Delta W / \Delta t$  the change in weight during the specified time interval (kg/s),  $r$  the radius (m) of the capillary and  $\rho$  the density of the electrolyte (kg/m<sup>3</sup>).

*On-line determination of EOF using a post-column detector.* In this method a constant reference flow is used in which a strong UV-absorbing marker is dissolved. This flow is mixed with the main flow (= EOF). By measuring the UV absorption in this mixed flow, information can be obtained about the ratio between the reference flow and the main flow. Because the reference flow is constant and known, the flow-rate of the main flow can also be calculated.

As a liquid pump for the reference flow, both a mechanical (Fig. 2) and an electroendosmotic (Fig. 3) pump can be used. Gas bubbles formed when using an "EOF" pump can be trapped, if they interfere with the main flow and stop the EOF (current switch-off), in different ways [e.g., bubble trap (Fig. 3, III)]. The result of the

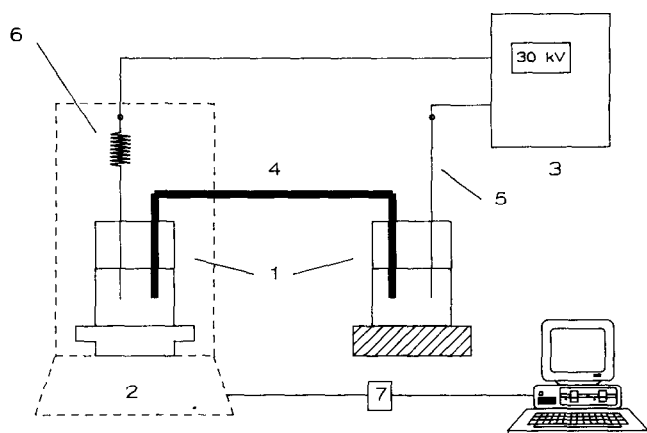


Fig. 1. Schematic representation of the on-line determination of the EOF by weighing. 1 = Electrolyte vessels; 2 = balance; 3 = high-voltage power supply; 4 = capillary; 5 = platinum electrode; 6 = balanced electrode; 7 = interface.

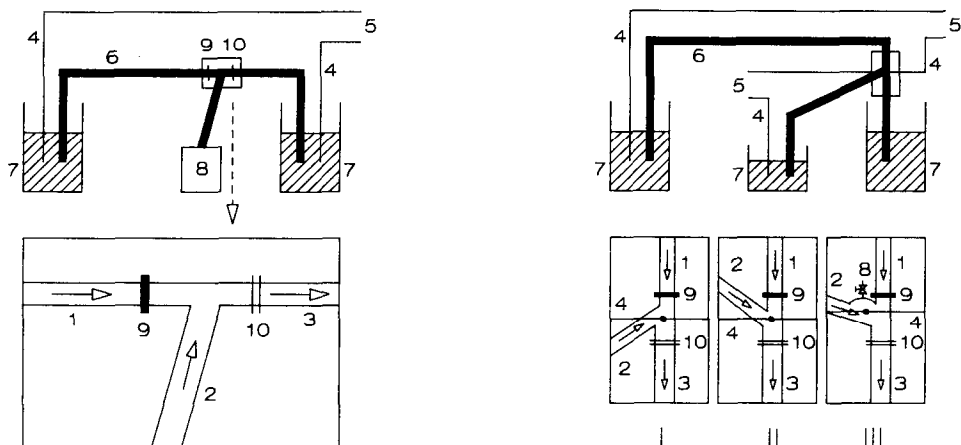


Fig. 2. Schematic representation of the on-line determination of the EOF using a post-column detector and a mechanical pump. 1 = Main flow; 2 = reference flow; 3 = mixed flow; 4 = electrodes; 5 = high-voltage power supply; 6 = capillary; 7 = electrolyte vessels; 8 = mechanical pump; 9 = analytical detector; 10 = post-column detector.

Fig. 3. Schematic representation of the on-line determination of the EOF using a post-column detector and an EOF pump. 1 = Main flow; 2 = reference flow; 3 = mixed flow; 4 = electrodes; 5 = high-voltage power supply; 6 = capillary; 7 = electrolyte vessels; 8 = bubble trap; 9 = analytical detector; 10 = post-column detector.

on-line measurement of the EOF is registered by a computer, to be used in the control unit.

*The control unit*

Direct control of the EOF can be achieved by coupling the computer to the high-voltage power supply (e.g., Gamma HV50000/1.25; Gamma High Voltage Research, Mt. Vernon, NY, U.S.A.). A change in the EOF can then be corrected by decreasing or increasing the voltage applied. Indirect control can be achieved by adjusting the speed of the recorder or by correcting the results, after the experiment, for the values of the EOF (Fig. 4).

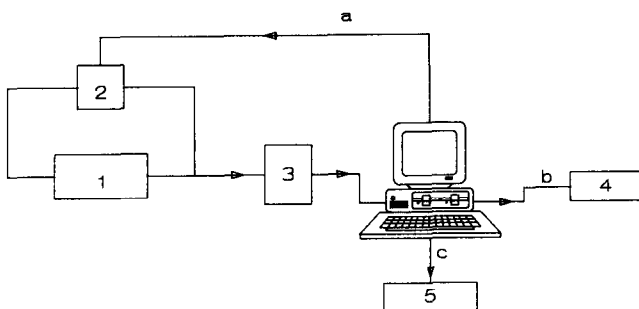


Fig. 4. Schematic representation of the control system. 1 = Separation compartment; 2 = high-voltage power supply; 3 = on-line measurement of the EOF; 4 = recorder; 5 = printer/screen. a = Direct control; b = indirect control by adjusting the speed of the recorder; c = indirect control by recalculation of the results after the experiment.

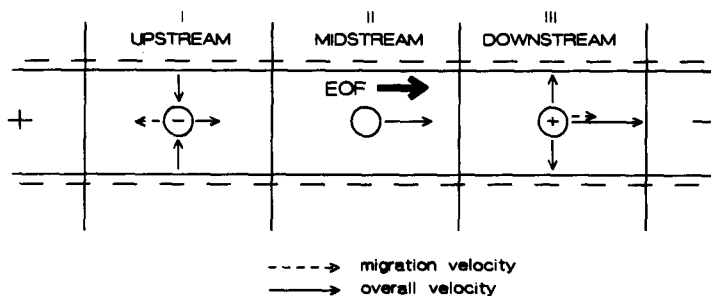


Fig. 5. Representation of the simultaneous separation of anions, cations and non-ionic components. The wall carries a negative charge. Repulsion of the anions in the upstream section and attraction of the cations in the downstream section is obtained.

### *Advantages of the control system*

Without an on-line measurement and control system, the EOF may vary under the influence of, *e.g.*, temperature or surface-active components, between two experiments, or even during one experiment. This leads to poor reproducibility of the results. The first advantage of the control system is an improvement in reproducibility for both electrophoretic and electro-chromatographic separations. In addition on-line control of the EOF also offers other possibilities for both CEC and CZE.

In CEC, the EOF can now be used as a reproducible pump, which can compete with, or even is better than, a mechanical pump. The advantage of using an "EOF" pump is its almost flat velocity profile. Consequently, the peak broadening is reduced and the separation is of better quality. In CZE, this control system for the EOF offers the possibility of a so-called "upstream" separation (Fig. 5, I). In this instance the separation of, *e.g.*, anions is performed while the EOF is flowing in the opposite direction. The combination of the increase in separation time and the repulsion between the ions and the capillary wall results in smaller plate heights and hence better resolution.

A final advantage of the control system is the possibility of combining both CZE and CEC techniques for the simultaneous separation of ionic and non-ionic components using only one detector. In Fig. 5 a difference is made between the "upstream" separation (discussed in the previous example), the "downstream" separation and "midstream" separation. In a "downstream" separation, non-ideal separation conditions can arise because of interactions between the wall and sample components and the decrease in separation time. The term "midstream" separation is used for the chromatographic separation of the non-ionic components; moreover, stationary phases can be used.

In this study the EOF has been measured<sup>12</sup>. The regulation of EOF and its contribution to the separation is under investigation.

### CONCLUSIONS

The vital condition of using EOF in both CEC and CZE is the possibility of an on-line measurement and control system. Such a control system not only improves the quality of CZE and CEC, but also adds some new possibilities to both techniques

such as the simultaneous separation of anions, cations and non-ionic components, using only one detector.

## REFERENCES

1. A. J. Rutgers, M. de Smet and W. Rigole, in H. van Olphen and K. J. Mysels, *Physical Chemistry: Enriching Topics from Colloid and Surface Sciences*, Theorex, La Jolla, CA, 1975.
2. R. J. Hunter, *Zeta Potential in Colloid Science*, Academic Press, London, 1981.
3. J. T. Davies and E. K. Rideal, *Interfacial Phenomena*, Academic Press, New York, 2nd ed., 1963.
4. A. W. Adamson, *Physical Chemistry*, Interscience, New York, 2nd ed., 1967.
5. F. E. P. Mikkers, F. M. Everaerts and Th. P. E. M. Verheggen, *J. Chromatogr.*, 169 (1979) 11.
6. J. E. Jorgenson and K. D. Lukacs, *Anal. Chem.*, 53 (1981) 1298.
7. H. H. Lauer and D. McManigill, *Anal. Chem.*, 58 (1986) 166.
8. J. H. Knox and I. H. Grant, *Chromatographia*, 24 (1987) 135.
9. V. Pretorius, B. J. Hopkins and J. D. Schieke, *J. Chromatogr.*, 99 (1974) 23.
10. T. Tsuda, K. Nomura and G. Nakagawa, *J. Chromatogr.*, 248 (1982) 241.
11. A. S. Cohen and B. L. Karger, *J. Chromatogr.*, 397 (1987) 409.
12. A. A. A. M. van de Goor, B. J. Wanders and F. M. Everaerts, *J. Chromatogr.*, 470 (1989) 95.