ANALYTICA CHIMICA ACTA 619 (2008) 115–121

available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/aca

In-channel simplified decoupler with renewable electrochemical detection for microchip capillary electrophoresis

*Kuan-Wen Lin, Yan-Kai Huang, Hsiu-Li Su, You-Zung Hsieh**

Department of Applied Chemistry, National Chiao Tung University, 1001 Ta Hsueh Road, Hsinchu 300, Taiwan

ARTICLE INFO

Article history:
Received 4 December 2007
Received in revised form
25 February 2008
Accepted 26 February 2008
Published on line 6 March 2008

Keywords:
Microchip capillary electrophoresis
Electrochemical detection
Decoupler
Renewable electrode

ABSTRACT

Electrochemical (EC) detection is comparable to fluorescence detection in that it is simple to perform, economical, and highly sensitive. In this study, we used replica molding to fabricate a PDMS microchip for microchip capillary electrophoresis (CE). A decoupler electrode and a working electrode were implanted into the PDMS chip during the molding process to prevent leakage into the electrode channel. The working electrode could be renewed readily through its slight withdrawal (ca. 3 mm) from the PDMS; its detection ability was highly reproducible in the microchip CE-EC system. The relative standard deviation (R.S.D.) of the detecting current for the renewed working electrode was 1.2% ($n = 5$). The calibration curves were linear for both dopamine and catechol analytes over the concentration range 10–1000 μM, with coefficients of determination (R^2) of 0.999 and 0.976, respectively. The number of theoretical plates (N/m) for these analytes was greater than 133,000.

1. Introduction

Microchip capillary electrophoresis (CE) is a useful separation technique that enables rapid detection of analytes within small sample volumes. Since the introduction of miniaturized CE systems over a decade ago [1], microchip CE has become increasingly popular, especially for high-throughput analyses, because of its low consumption of reagents, low generation of waste, high disposability and portability, the ability to integrate pre-treatment and post-separation processes, and the ability to perform multiplexed analysis [2–6]. Although laser-induced fluorescence (LIF) is the most commonly utilized optical detection method for analytes in miniaturized separation channels [7–9], high sensitivity is usually achieved only when the analyte is derivatized prior to analysis. Unlike optical methods, the performance of electrochemical (EC) detection systems does not decline upon miniaturizing the electrodes [10]; in addition, EC allows opaque materials to be used in the microchips [11]. Thus, EC detection for microchip CE analyses is becoming increasingly popular [3,4,12]. Of the three EC methods – amperometry [13,14], conductimetry [15,16], and voltammetry [17] – that have been applied to microchip CE detection, amperometric detection is the most popular. Examples of microchip CE-EC applications are found in enzyme assays, immunoassays, neurotransmitter analysis, clinical diagnostics, and environmental monitoring [18–21]. Many of the methods used to fabricate microfluidic channels can also be employed to construct microchip CE-EC systems. A fully integrated microchip containing electrodes for both electrophoresis and EC detection has been demonstrated [22]. With the ability to miniaturize power supplies and EC analyzers [23,24], it is possible to envision a complete micro-total-analysis system (μ-TAS).

* *Corresponding author*. Tel.: +886 3 5731785; fax: +886 3 5723764.
E-mail address: yzhsieh@mail.nctu.edu.tw (Y.-Z. Hsieh).
0003-2670/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.aca.2008.02.062

There are three main approaches to integrating EC with microchip CE: end-channel, in-channel, and off-channel detection [25]. For end-channel detection, the working electrode is positioned in the exit of microchannel. The effect of the separation field on the working electrode is minimized through use of extremely low separation currents. Although the detection noise due to the separation current decreases when the working electrode is positioned further from the microchannel outlet, the detection sensitivity also decreases as a result of the loss of analytes through diffusion in the detection cell [26]. In-channel and off-channel detection systems can be employed to prevent such loss of analytes, but interference from the high electric field and electrophoretic current on the EC measurement can be problematic, leading to a larger EC background current and a shifted working electrode potential [25]. An in-channel amperometric detection system lacking a decoupler has been developed; it uses an electrically isolated potentiostat [24]. In the off-channel arrangement, the separation potential must be grounded before the analyte reaches the microchannel outlet to eliminate interference from the separated electric field and to protect the EC detector from damage resulting from surges in current. A decoupler placed in front of the working electrode can be used to protect the EC detection system from interference [27–32]. Currently, several types of decoupler have been fabricated, including thin film [28–30] and microwire [31] electrodes. A solid Pd film integrated directly into the CE microchip across the separation channel has been prepared using microfabrication techniques; it replaced the decoupler in a joint connection-type system [28]. The reason why Pd metal was adopted is that platinum (Pt)-group metals, such as Pd and Pt, effectively reduce and absorb hydrogen ions. Because molecular hydrogen diffuses faster on a Pd surface, it is eliminated from the Pd decoupler before the development of hydrogen bubbles in the electroosmotic flow (EOF), maintaining the efficiency of the decoupler [28,29]. A Pd microelectrode wire decoupler and metal wire working electrode have been integrated within a microchip CE-EC system to eliminate the microfabrication steps used to construct the electrodes [31].

The fouling of metal working electrodes is a serious concern affecting microchip CE-EC [33]. Inactivation of electrodes upon repeated injection or varying analyte concentrations affects the sensitivity, limit of detection, and reproducibility of microchips. Although surface regeneration of an electrode can improve its performance, an optimized cleaning procedure applied to a specific electrode might not be applicable to routine retuning of all electrodes.

Most CE microchips have been fabricated from glass or quartz because such materials are suitable for optical detection and readily generate EOFs. Unfortunately, the fabrication of glass substrate microchips requires clean-room processing and high-temperature thermal bonding; thus, it is difficult to perform in most general laboratories [34]. A more amenable approach is the use of polymers, such as poly(methyl methacrylate) (PMMA), polycarbonate, polyethylene, and poly(dimethylsiloxane) (PDMS), rather than glass or silicon, to form CE microfluidic channels [35]. Of these polymers, PDMS has been applied most widely to microchip fabrication because it exhibits several favorable characteristics: (i) its optical transparency down to 230 nm makes it suitable for optical detection; (ii) it cures at low temperatures so that molding is readily replicated through a process of master formation, prototyping, and soft lithography; (iii) it can be sealed reversibly to itself and other materials with a clear smooth surface at room temperature through van der Waals contacts; (iv) it can be sealed permanently to itself and glass; and (v) its surface chemistry can be controlled to form an EOF using plasma techniques [36]. The EOF of the oxidized PDMS channel is directed toward the cathode; it is caused by the hydrolysis of siloxane bonds in the oxidized layer [37]. Microchip CE utilizing a PDMS slice combined with a glass substrate was first developed by Effenhauser [38]; since then, many applications of PDMS in microchip CE-EC and microfluidic channel design have been reported [39].

In this paper, we report a simple method for integrating a decoupler and a working electrode into PDMS-based microchip CE-EC devices. A Pt wire electrode serving as a decoupler and a Cu working electrode were incorporated into a PDMS sheet. We employed catechol and dopamine as analytes to demonstrate the separation efficiency of the microchip CE-EC system and to evaluate the performance of the renewable working electrode. We believe that the extended reuse of such renewable working electrodes coupled with these simple microchip fabrication techniques will increase the usability of microchip CE-EC systems.

2. Experimental

2.1. Reagents

The Sylgard 184 prepolymer and its curing agent were purchased from Dow Corning (Midland, MI, USA). Disodium hydrogen phosphate, sodium dihydrogen phosphate, sodium hydroxide, sodium dodecyl sulfate (SDS), and other chemicals for electrophoresis were purchased from Fluka (Buchs, Switzerland). Catechol and dopamine were purchased from Sigma (St. Louis, MO, USA). Stock solutions (10 mM) of catechol and dopamine standards were prepared in acidic solution and used within 6 h. Analytical-grade isopropanol and acetone, which were used to wash the PDMS chips and patterned wafers, were purchased from Union Chemical Work (Hsinchu, Taiwan).

2.2. Apparatus

The patterned silicon wafer used to mold the PDMS microchannel was designed using standard computer software (AutoCAD 2000) and prepared at the Industrial Technology Research Institute of Taiwan (Hsinchu, Taiwan). The channel dimensions on the silicon wafer were 50 μm (wide) by 50 μm (deep). The diameter of the Pt wire of the decoupler was 500 μm. The Cu wire of the working electrode, having a diameter of 200 μm, was obtained as a gift from the Yeou-Chuen Wire Co. (Taoyuan, Taiwan). The purity of the Pt and Cu wires was 99.99%. The reference electrode was MF-2052 Ag/AgCl (Bioanalytical System, Baltimore, MD, USA). The reference and auxiliary electrodes were placed in the buffer waste reservoir.

The detection system was an 802b EC analyzer (CH Instruments, Austin, TX, USA), coupled to the working, auxiliary, and reference electrodes through sockets. An MP-5000-250P high-voltage power supply system (Major Science, Taipei, Taiwan) having a programmable adding voltage system and an adjustable voltage range from 0 to +5 kV was used to effect CE separation on the microchip.

2.3. *In-column electrode setting*

The procedures used to place the metal wires and assemble the microchip are illustrated in Fig. 1(a); the completed microchip is displayed in Fig. 1(b). The Pt and Cu wires were fixed precisely on the patterned silica wafer, and then PDMS was poured into the mold and cured; the decoupler and working electrode were aligned horizontally on the PDMS. The distances between the center of the decoupler and the end of microchannel and the center of the working electrode and the end of microchannel were controlled at 1000 and 200 μm, respectively, and the active surface areas were ca. 750 and 250 μm^2, respectively, as indicated in Fig. 1(b). The distance between the decoupler and the working electrode was used to maximize the separation efficiency and to minimize the interference from the separation field. The distance between the working electrode and the end of the microchannel was minimized to maintain a stable electrode potential toward the reference electrode. The microchip was separated into two parts: PDMS, containing the electrodes and microchannels, and bare sodium glass (each ca. 3 cm × 9 cm × 0.3 cm). Both portions were cleaned sequentially using neutral detergent, acetone, isopropanol, and deionized water for 15 min each, exposed to air plasma for 1 min, and then sealed together. The surfaces of the working electrodes were very smooth; therefore, when they became polluted by analytes, they could be drawn out and easily renewed. The procedures used to renew the working electrode are illustrated in Fig. 2.

2.4. *Electrophoresis and detection*

Prior to use, the microchannels of the PDMS chip were rinsed with deionized water and running buffer solution (5 min each). The electrophoretic separation medium (usually 5 mM phosphate buffer solution, pH 7.4) was prepared afresh daily using deionized water. The Cu wire working electrode was placed near the buffer waste reservoir. Amperometric detection was performed using an 802b EC analyzer (CH Instruments, Austin, TX, USA) operated in the "amperometric i–t curve" mode. The injection volume was controlled by the length of the patterned distance between the sample channel, as indicated in Fig. 1(b); the injection length using different sample reservoirs (S1, S2, and S3) were 300 (S1, sample inlet; S2, sample waste), 550 (S2, sample inlet; S3, sample waste), and 800 (S1, sample inlet; S3, sample waste) μm, respectively. The reservoirs were filled with running buffer and the sample solution and a sample injection potential of 100 V cm^{-1} was then applied for 15 s. Separation was then initiated by switching to different voltages across the separation channel. Sample injections and separations were performed after stabilization of the baseline.

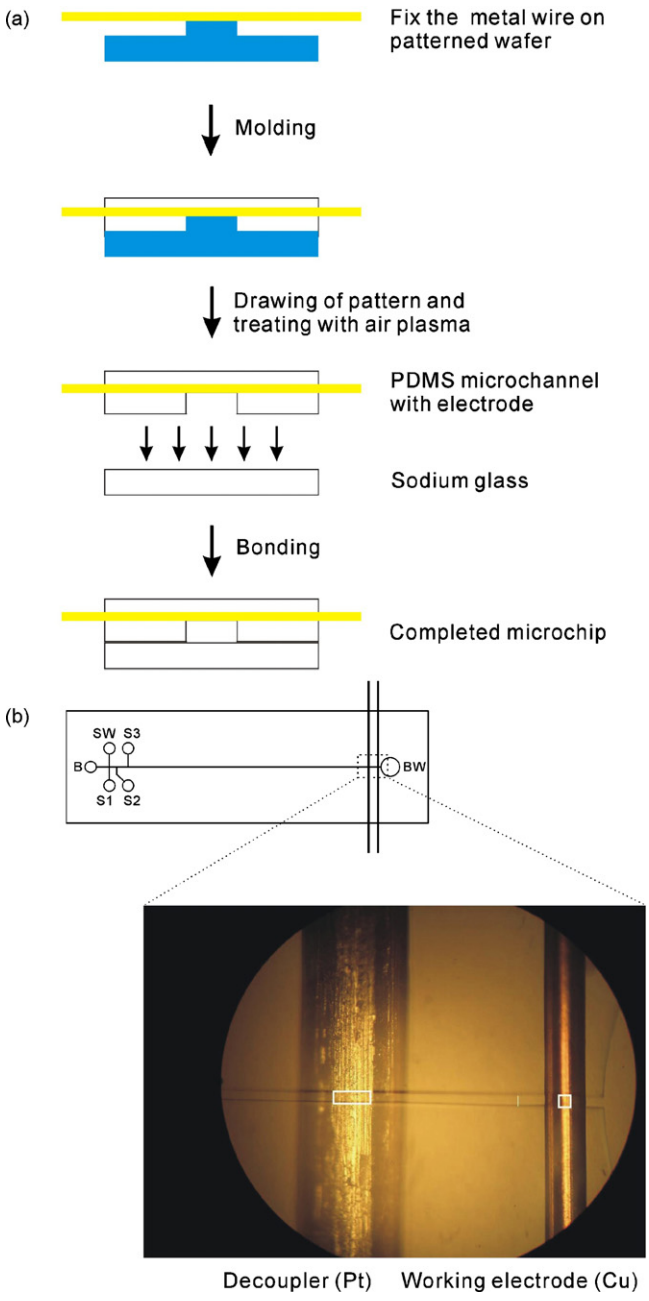

Fig. 1 – (a) Procedure for the fabrication of the in-channel decoupler/renewable electrode microchip CE-EC system. (b) Schematic representation of the microchip CE-EC system. Reservoir functions: B, buffer solution; BW, buffer waste; S1, S2, and S3, sample inlets; SW, sample waste. The distances from the center of the decoupler to the end of the microchannel and the center of the working electrode to the end of the microchannel were 1000 and 200 μm, respectively. The diameters of the decoupler and working electrode were 500 and 200 μm, respectively. The white rectangles mark the contact areas of the decoupler and electrode in the channel.

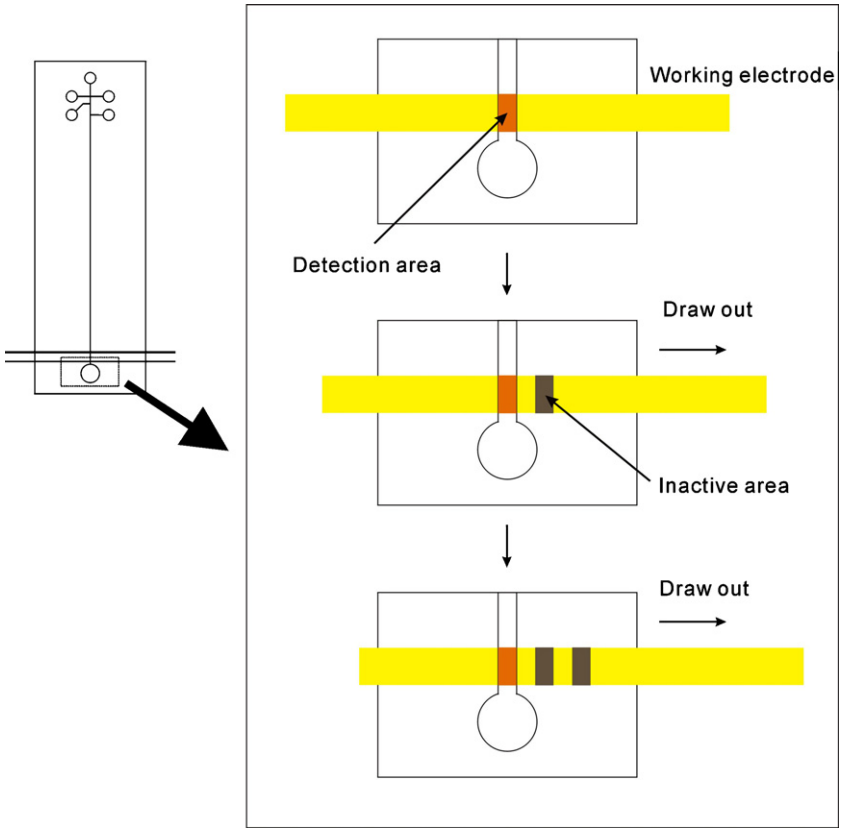

Fig. 2 – Renewing the working electrode. The electrode was withdrawn by ca. 3 mm each time.

3. Results and discussion

3.1. *Hydrodynamic voltammetry*

The presence of an in-channel decoupler and off-channel working electrode characterizes an EC detector that improves amperometric detection. We obtained hydrodynamic voltammograms (HDVs) to illustrate the enhanced electrocatalytic properties of copper electrodes, making it possible to detect analytes at relatively low potentials. Fig. 3 depicts typical HDVs for the oxidations of both 125 μM dopamine and catechol at the off-channel Cu detector. The curves were developed pointwise, in steps of 0.1 V (vs. Ag/AgCl), over the range of −0.2 to 0.4 V using a separation voltage of 150 V cm^{-1}. The voltammograms indicate that the oxidative current was effectively obtained when using the in-channel decoupler to ground the separation voltage. Although increasing the separation field decreased the separation time, the highest signal-to-noise detection current was obtained at a separation voltage of 150 V cm^{-1}. For both analytes, the oxidation began at the 0.0 V potential of the Cu electrode. We observed distinct oxidative currents at 0.0 and 0.2 V in the range of tested potentials. Although the highest oxidative current appeared at 0.2 V, a concurrent dramatic increase in the baseline noise occurred at this potential. Therefore, for all subsequent amperometric detection experiments we employed a constant potential of 0.0 V, which provided the highest signal-to-noise detection current.

3.2. *SDS dynamic modification*

Manipulating the EOF by adjusting the surface state of PDMS-based microchannels is critical to enhancing the repro-

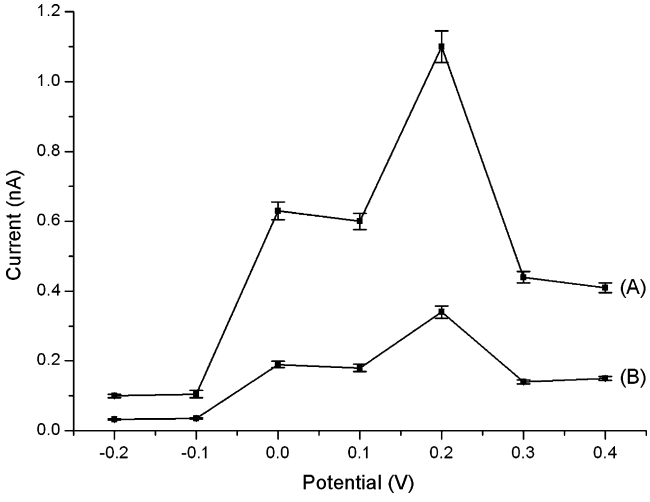

Fig. 3 – Hydrodynamic voltammograms for (A) 125 μM dopamine and (B) 125 μM catechol at the Cu working electrode, constructed through stepwise (0.1 V) increases in the detection potential (vs. Ag/AgCl). Each value denotes mean ± S.D. of five replicates. Conditions: phosphate buffer solution containing 0.8 mM SDS (5 mM, pH 7.4); separation voltage, 150 V cm^{-1}; injection voltage, 400 V cm^{-1}; injection time, 5 s.

Table 1 – Influence of SDS on the migration time and detection current for catechol

	Operation time (h)				Average current (10^{-10} A)[b]	R.S.D. (%)[c]
	0	1	2	3		
t_M (s)[a]						
Without SDS	44.8	46.0	46.3	43.9	1.33	2.4
With SDS	45.0	45.8	45.8	46.0	2.83	1.0

[a] Migration time.
[b] Average current of 125 μM catechol during 3-h operation ($n = 4$).
[c] R.S.D. of migration time ($n = 4$).

ducibility of such microchips. Dynamic modification through the addition of surfactants in the running buffer is a simple means of adjusting the EOF [40]. In this study we varied the concentration of SDS to determine its effect on separation. Table 1 summarizes the experimental results in terms of the migration time and detection current for catechol in the presence and absence of 0.8 mM SDS. Adding SDS to the buffer improved the R.S.D. of the migration time; compared with the results obtained without adding SDS, the detection signal increased 2.13-fold under a similar noise level. According to these results, adding SDS to the buffer enhanced the stability of PDMS microchip CE, presumably because the absorbed surfactant improved the interactions between the analytes and the working electrode or the channel surface. Adding too much SDS, however, increased the separation current and led to bubbles of H_2 appearing at the electrode. Therefore, for subsequent experiments we chose to use 0.8 mM SDS as the dynamic modifying agent.

3.3. *Performance of microchip CE-EC*

The detected EC signal usually decreases after operating a microchip CE-EC system several hours. Inactivation of the Cu working electrode is an unavoidable problem for EC detection, especially for microchip CE-EC. The degree of inactivation of the electrode is related to the degree of continued use of the microchip. Even if the electrode could be reactivated, its sensitivity would still decrease after several reactivation cycles. As a potential solution to this problem, Fig. 2 illustrates our simple concept for renewing the electrode: the off-channel Cu electrode is slowly drawn out to a distance of 3 mm to renew the detection surface. Theoretically, hydrogen gas bubbles will form after the Pt electrode is saturated with hydrogen atoms. In this system, it took more than 20 min to generate bub-

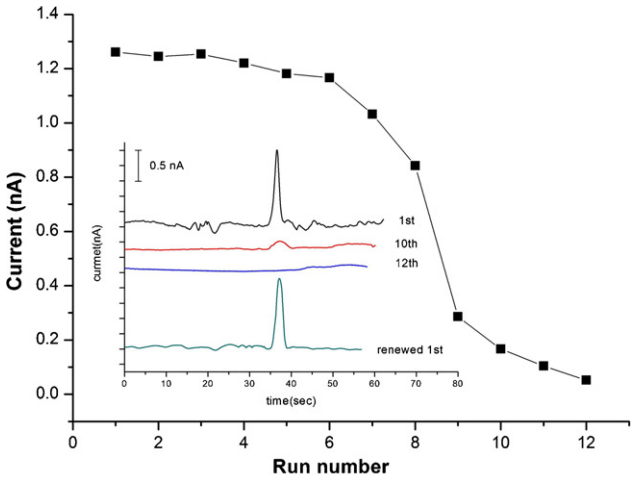

Fig. 4 – Using the working electrode to detect 250 μM dopamine during 12 sequential analyses. Electropherograms displaying the first, 10th, and 12th runs, and the first run using the renewed electrode. The injection length was 800 μm; all other conditions were the same as those described for Fig. 3.

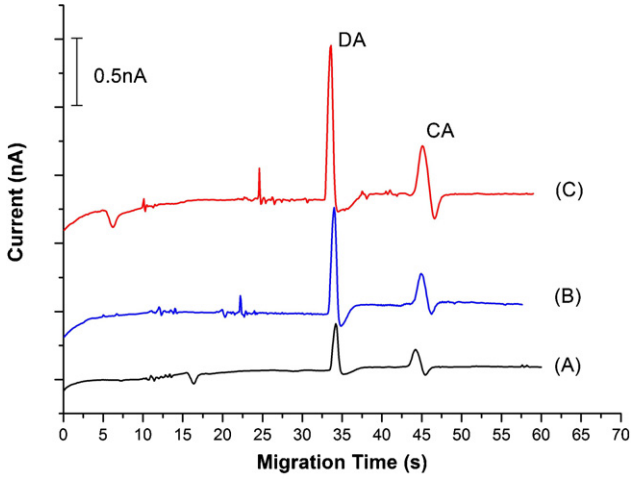

Fig. 5 – Electropherograms obtained using injection lengths of (A) 300 μm, (B) 550 μm, and (C) 800 μm. All other conditions were the same as those described for Fig. 4.

Table 2 – Performance of the renewable working electrode for the detection of dopamine

	Average current (nA)	R.S.D. (%)[a]	Average migration time (s)	R.S.D. (%)[b]
First run[c]	1.255	1.2	35.3	1.1
Five runs[d]	1.232	2.6	36.1	2.9

[a] R.S.D. of average current.
[b] R.S.D. of average migration time.
[c] First run of each renewed working electrode ($n = 5$).
[d] Five consecutive runs using the same detection area.

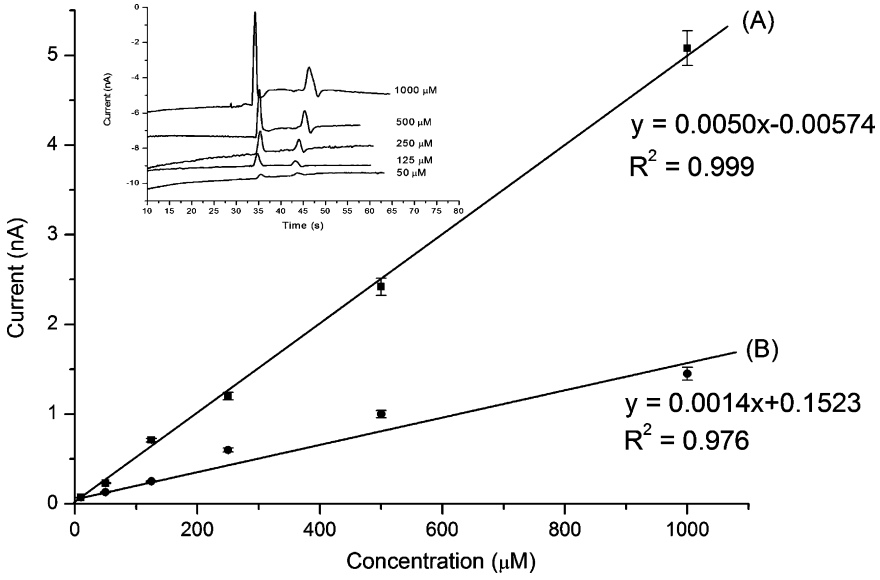

Fig. 6 – Calibration curves of (A) dopamine and (B) catechol over the concentration range from 10 to 1000 μM. Each value denotes mean ± S.D. of five replicates. The conditions were the same as those described for Fig. 4.

bles under an electric field of 150 V cm^{-1}. The bubbles in the channel were readily eliminated through flushing with fresh running buffer.

Fig. 4 displays the detecting currents obtained during 12 analyses without renewing the detection area of the electrode. The detecting current decreased significantly after the eighth run. After renewing the working electrode, the electropherogram indicates that the sensitivity was similar to that of the original detecting zone. Table 2 lists the average values of the detection current, migration time, and R.S.D. for the performance of this renewable electrode. The values of R.S.D. of the detecting current and migration time were both less than 1.2% for the first runs using the renewed electrode. These values are even better than those obtained from five consecutive runs using the same detection area. Because of the tight fit of the working electrode and the elasticity of the PDMS, we did not observe any leakage at the site at which the channel crossed the working electrode. Because PDMS possesses low chemical reactivity, it is safe to assume that the properties of the Cu electrode remain unchanged within the polymeric material. This simple design for a renewable working electrode provides exceptional performance while extending the working life of the microchip CE-EC system.

The combination of an in-channel decoupler and a renewable off-channel working electrode results in a stable, rapid, highly sensitive, and highly reproducible microchip system. Fig. 5 displays electropherograms of mixtures of dopamine and catechol analyzed using injection lengths of 300, 550, and 800 μm. The detection currents increased upon increasing the injection length; the heights of the analyte peaks increased accordingly, but maintained similar peak widths, presumably because of the presence of the in-channel decoupler and the dynamically modified separation channel. Fig. 6 presents calibration curves and electropherograms for various concentrations of the analytes (dopamine: 10–1000 μM; catechol: 50–1000 μM). The detection limits for dopamine and catechol were 2.8 and 12.7 μM, respectively. Using this off-channel detection system, the time required for the analytes to flow through the working electrode was shorter and the detection limit higher than those obtained using the end-channel detector. The theoretical plate numbers (*N*/m) for dopamine and catechol were 133,000 and 160,000, respectively, values that are ca. 3–10-fold larger than those obtained using end-channel and/or dual-channel detection methods [41,42]. Therefore, our design for an in-channel simplified decoupler coupled with an off-channel detector increased the separation efficiency while minimizing peak broadening.

In a previous report, prearranged channels were used to fix the decoupler and working electrode, which were sealed with epoxy to prevent leakage into electrode channels [43]. In this present study, we incorporated the decoupler and working electrode into the PDMS chip during the molding process. This design prevents leakage into the electrode channels, as evidenced by the lack of peak tailing. Based on this prototype, our future efforts will be aimed at the application of a Cu working electrode with appropriate modification to the monitoring of a wider range of important analytes.

4. Conclusion

We have demonstrated the applicability of off-channel EC detection for microchip CE. A simultaneously integrated decoupler and off-channel renewable working electrode is relatively easy to fabricate in a microchip CE-EC system. Using the in-channel decoupler, the separation current remained stable even under high field strength. Off-channel detection for microchip CE-EC systems provided several benefits to the performance of the microanalytical system, e.g., noise at the detector was less than 0.01 nA and detection with the renewable working electrode was highly reproducible (R.S.D. = 1.2%). We also found that the renewed working electrode provided

a stable potential when performing amperometric detection. When using the renewable electrode, the lifetime of each active CE-EC microchip increased substantially. Further development on the working electrode through coating with a suitable nano-material to improve its sensitivity will enhance its practical applications. We hope that this approach will advance the development and miniaturization of lab-on-a-chip CE-EC systems.

Acknowledgement

This study was supported by grant number NSC 95-2113-M-009-033-MY3 from the National Science Council of Taiwan.

REFERENCES

[1] A. Manz, N. Graber, H.M. Widmer, Sens. Actuator B 1 (1990) 244.
[2] M. Freemantle, Chem. Eng. News 77 (1999) 27.
[3] N.A. Lacher, K.E. Garrison, R.S. Martin, S.M. Lunte, Electrophoresis 22 (2001) 2526.
[4] W.R. Vandaveer IV, S.A. Pasas, R.S. Martin, S.M. Lunte, Electrophoresis 23 (2002) 3667.
[5] M.A. Unger, H.P. Chou, T. Thorsen, A. Scherer, S.R. Quake, Science 288 (2000) 113.
[6] J.M. Ng, I. Gitlin, A.D. Stroock, G.M. Whitesides, Electrophoresis 23 (2002) 3461.
[7] V. Heleg-Shabtai, N. Gratziany, Z. Liron, Anal. Chim. Acta 571 (2006) 228.
[8] Y. Xiao, X.D. Yu, K. Wang, J.J. Xu, J. Huang, H.Y. Chen, Talanta 71 (2007) 2048.
[9] Q. Lu, C.L. Copper, G.E. Collins, Anal. Chim. Acta 572 (2006) 205.
[10] R.M. Wightman, Anal. Chem. 53 (1981) 1125A.
[11] C.S. Henry, M. Zhong, S.M. Lunte, M. Kim, H. Bau, J. Santiago, Anal. Commun. 36 (1999) 305.
[12] M.A. Schwarz, P.C. Hauser, Lab Chip 1 (2001) 1.
[13] J. Wang, G. Chen, M. Wang, M.P. Chatrathi, Analyst 129 (2004) 512.
[14] J. Wang, M.P. Chatrathi, B. Tian, Anal. Chem. 72 (2000) 5774.
[15] J. Lichtenberg, N.F. de Rooij, E. Verpoorte, Electrophoresis 23 (2002) 3769.
[16] A. Berthold, F. Laugere, H. Schellevis, C.R. de Boer, M. Laros, R.M. Guijt, P.M. Sarro, M.J. Vellekoop, Electrophoresis 23 (2002) 3511.
[17] J. Wang, R. Polsky, B. Tian, M.P. Chatrathi, Anal. Chem. 72 (2000) 5285.
[18] J. Wang, A. Ibáñez, M.P. Chatrathi, A. Escarpa, Anal. Chem. 73 (2001) 5323.
[19] M. Vlćková, M.A. Schwarz, J. Chromatogr. A 1142 (2007) 214.
[20] J. Wang, M.P. Chatrathi, G.E. Collins, Anal. Chim. Acta 585 (2007) 11.
[21] G. Chen, Y. Lin, J. Wang, Talanta 68 (2006) 497.
[22] R.P. Baldwin, T.J.J. Roussel, M.M. Crain, V. Bathlagunda, D.J. Jackson, J. Gullapalli, J.A. Conklin, R. Pai, J.F. Naber, K.M. Walsh, R.S. Keynton, Anal. Chem. 74 (2002) 3690.
[23] D.J. Jackson, J.F. Naber, T.J.J. Roussel, M.M. Crain, K.M. Walsh, R.S. Keynton, R.P. Baldwin, Anal. Chem. 75 (2003) 3643.
[24] R.S. Martin, K.L. Ratzlaff, B.H. Huynh, S.M. Lunte, Anal. Chem. 74 (2002) 1136.
[25] J.J. Xu, A.J. Wang, H.Y. Chen, Trends Anal. Chem. 26 (2007) 125.
[26] P. Ertl, C.A. Emrich, P. Singhal, R.A. Mathies, Anal. Chem. 76 (2004) 3749.
[27] D.M. Osbourn, C.E. Lunte, Anal. Chem. 75 (2003) 2710.
[28] D.C. Chen, F.L. Hsu, D.Z. Zhan, C.h. Chen, Anal. Chem. 73 (2001) 758.
[29] N.A. Lacher, S.M. Lunte, R.S. Martin, Anal. Chem. 76 (2004) 2482.
[30] D.M. Tsai, K.W. Lin, J.M. Zen, H.Y. Chen, R.H. Hong, Electrophoresis 26 (2005) 3007.
[31] J.A. Vickers, C.S. Henry, Electrophoresis 26 (2005) 4641.
[32] M.L. Kovarik, M.W. Li, R.S. Martin, Electrophoresis 26 (2005) 202.
[33] D.P. Manica, Y. Mitsumori, A.G. Ewing, Anal. Chem. 75 (2003) 4572.
[34] J.P. Kutter, Trends Anal. Chem. 19 (2000) 352.
[35] R.S. Martin, A.J. Gawron, S.M. Lunte, C.S. Henry, Anal. Chem. 72 (2000) 3196.
[36] J.C. McDonald, D.C. Duffy, J.R. Anderson, D.T. Chiu, H. Wu, O.J.A. Schueller, G.M. Whitesides, Electrophoresis 21 (2000) 27.
[37] D.C. Duffy, J.C. McDonald, O.J.A. Schueller, G.M. Whitesides, Anal. Chem. 70 (1998) 4974.
[38] C.S. Effenhauser, G.J.M. Bruin, A. Paulus, M. Ehrat, Anal. Chem. 69 (1997) 3451.
[39] M. Pumera, A. Merkoci, S. Alegret, Trends Anal. Chem. 25 (2006) 219.
[40] C.D. García, C.S. Henry, Anal. Chim. Acta 508 (2004) 1.
[41] A.T. Woolley, K. Lao, A.N. Glazer, R.A. Mathies, Anal. Chem. 70 (1998) 684.
[42] C. Chen, J.H. Hahn, Anal. Chem. 79 (2007) 7182.
[43] Y. Liu, J.A. Vickers, C.S. Henry, Anal. Chem. 76 (2004) 1513.