Journal of Chromatography A, 1416 (2015) 94–102

Contents lists available at ScienceDirect

Journal of Chromatography A

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

A facile and versatile approach for controlling electroosmotic flow in capillary electrophoresis via mussel inspired polydopamine/polyethyleneimine co-deposition

Qifeng Fu, Xiuju Li, Qihui Zhang, Fengqing Yang, Weili Wei, Zhining Xia*

School of Chemistry and Chemical Engineering, Chongqing University, Chongqing 400030, China

ARTICLE INFO

Article history:
Received 3 June 2015
Received in revised form 20 August 2015
Accepted 3 September 2015
Available online 6 September 2015

Keywords:
Capillary modification
Controllable electroosmotic flow
Polydopamine
Polyethyleneimine
Co-deposition

ABSTRACT

Electroosmotic flow (EOF), which reveals the charge property of capillary inner surface, has an important impact on the separation performance and reproducibility of capillary electrophoresis (CE). In this study, a novel, facile and versatile method to achieve diverse and controllable EOF in CE was reported based on the co-deposition of mussel-inspired polydopamine (PDA) and branched polyethyleneimine (PEI) on the capillary inner surface as the hybrid functional coating. After these PDA/PEI co-deposited columns were reinforced by the post-incubation of $FeCl_3$, various magnitude and direction of EOF in CE could be easily achieved by varying a number of preparation parameters, including the mass ratio of DA/PEI and the molecular weight of PEI (including PEI-600, PEI-1800, PEI-10000 and PEI-70000). The separation effectiveness and stability of the hybrid coated columns were verified by the analysis of aromatic acids and aniline derivatives. The results showed that the controllable and diverse EOF was important in enhancing the separation performance of the analytes. The baseline separation of all the five aromatic acids can be achieved in 7 min with high separation efficiency by using the PDA/PEI-600 co-deposited column with the mass ratio of 6:1. On the other hand, with the PDA/PEI-70000 co-deposited column with the mass ratio of 6:1, the aniline compounds were easily baseline separated within 10 min. By contrast, using the bare and PDA coated columns, the migration of the aromatic acids was very slow and the baseline separation of the aniline compounds cannot be obtained. Moreover, the co-deposited columns showed long lifetime and good stability. The relative standard deviations for intra-day, inter-day and capillary-to-capillary repeatability of the PDA/PEI-600 co-deposited column with the mass ratio of 6:1, which was reinforced by the post-incubation of $FeCl_3$, were all lower than 5%.

1. Introduction

Electroosmotic flow (EOF), an important electrokinetic phenomenon in capillary electrophoresis (CE), has an important impact on the separation performance and reproducibility of CE. Variations in the chemistry of the fused silica surface often result in insufficient EOF reproducibility, which in turn impedes the acceptance of CE [1,2]. On the other hand, the cathodic EOF provided by the bare silica capillary is not optimal for many CE applications. To achieve the optimal separations, control of EOF is critical in many cases [1,3]. For example, extremely rapid separation can be achieved if the electrophoretic mobility and EOF are in the same direction. Alternatively, high-resolution separation can be obtained when EOF is in the opposite direction of the analytes movement. Besides, the negatively charged silica surface is easily adsorbed by the positively charged analytes, especially the polycationic macromolecules such as proteins [1,2], which will decrease the separation performance of CE.

To control the EOF in CE, various coating approaches related to modification of the capillary surface have been presented, including covalent coating [1,4,5], dynamic coating [6–8], and physically adsorbed coating [9–11]. Covalent coating is a widely used modification strategy, which possesses good stability and long lifetime [1,12]. However, the tedious and time-consuming preparation process will increase the cost of a CE analysis. Alternatively, many charge modifiers such as surfactants [6] and functionalized nanoparticles [7,8] have been used to modify the capillary surface by dynamic coating. Nevertheless, the dynamic modification often needs regeneration or the charge modifiers as pseudostationary phases in the running buffer, which may affect on the separation and detection of analytes. Another modification protocol is associated with the deposition of charge modifiers on

---

* Corresponding author.
E-mail address: tcm_anal_cqu@163.com (Z. Xia).

http://dx.doi.org/10.1016/j.chroma.2015.09.014
0021-9673/© 2015 Elsevier B.V. All rights reserved.

the capillary surfaces using physical adsorption, which is easy to fabricate with low cost [9,12]. However, compared with covalent coating, physically adsorbed coating normally is less stable and has a shorter lifetime. Moreover, it is difficult to tailor the magnitude and direction of EOF over a wide range solely through a certain coating approaches. Consequently, the limited diversity and flexibility in tailoring EOF would hinder the possibility of employing these modification approaches for the separation of a wider range of analytes.

To address the challenge of controlling the magnitude or direction of EOF over a wide range, some capillary modification approaches have been presented. Lucy and co-workers proposed a dynamic coating strategy using the mixture of cationic and zwitterionic surfactants to control the amplitude of an anodic EOF (i.e. a reversed EOF from the cathode to the anode) via the alteration of relative concentration of both surfactants [13]. Afterwards, they further developed a surfactant bilayer/diblock polymer coating strategy to control the amplitude of an anodic EOF by varying the polyoxyethylene stearate chain length [14]. Similarly, Erny's group used the mixture of anionic and neutral surfactants to adjust the magnitude of a cathodic EOF [15]. However, these surfactant coatings have poor stability and the frequent regeneration is indispensable. Additionally, the EOF can only be varied in one direction and the available variation range of EOF is limited. Some new copolymers possessing different charge densities have been synthesized and coated on the capillary surfaces via the physical adsorption, which can control the direction and magnitude of the EOF in CE [16,17]. Nevertheless, the synthesis of these charge modifiers is not easy and costly.

Mussel-inspired surface chemistry on the polydopamine (PDA) has attracted extensive interest in different research fields [18,19], including CE. Based on the diverse properties of PDA such as strong adhesive property [18–20], metal ion chelation and redox activities [19,21], latent reactivity with many functionalized molecules [19,22], several studies on the preparation and application of PDA-based coatings in CE have been reported [23–27]. Yin and co-workers first reported the use of PDA as a stationary phase in open tubular capillary electrochromatography (OT-CEC) for the determination of auxins based on the strong adhesion of PDA [23]. Wang's group anchored antifouling amine-functionalized PEG on the capillary inner surface for the protein separation, which was based on the latent reactivity of PDA [24]. Furthermore, graphene or bovine serum albumin have been immobilized on the PDA coated capillary for the separation of alkylbenzenes or the enantioseparation of amino acid enantiomers [25,26]. Additionally, PDA also can be used to assist the biomineralization of hydroxyapatite as the stationary phase of OT-CEC for the separation of alkylbenzenes, phenols and amines [27]. On the other hand, because of the coexistence of amine and phenolic hydroxyl groups, it is possible that the zwitterionic property of PDA also can be utilized for surface charge modification to obtain diverse and controllable EOF in CE [19,28]. However, due to the limited molecular diversity of coating precursors, it is difficult to render PDA with diverse desired surface charge characters, a critical factor affecting the diversity of EOF, solely through dopamine (DA). Recently, the surface modification strategies involved in the co-deposition of PDA and some other functionalized molecules have been proposed [29–32], which can render the materials with various desired properties. Considering the variety of co-deposited functionalized molecules, this strategy has potency to expand the surface charge diversity of PDA-based materials by co-depositing various charge modifiers. However, to our knowledge, the potential use of hybrid PDA coatings to achieve diverse and controllable EOF in CE has not been reported.

In the present study, to reveal the potential use of co-deposited PDA-based materials to achieve diverse and controllable EOF in CE, a facile and versatile approach to fabricate the PDA/branched polyethyleneimine (PEI) co-deposited coatings in capillary inner surface with diverse surface charge features was proposed for the first time. The morphology and surface properties of these columns were characterized by field emission scanning electron microscopy (FESEM) and attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FT-IR). The existence of the cross-linking reaction between DA and PEI was verified by UV–visible and fluorescence spectroscopies. Furthermore, the different coating preparation parameters such as the post-incubation time of $FeCl_3$, the co-deposition time, the mass ratio of DA and PEI and the molecular weight of PEI were intensively studied and compared. Additionally, the stability and separation efficiency of the constructed PDA/PEI modified columns were verified by the separation of aromatic acids and aniline compounds.

2. Materials and methods

2.1. Reagents and chemicals

Dopamine hydrochloride, p-anisidine, p-toluidine, N,N-dimethylaniline and PEI (with different molecular weight of 600, 1800, 10,000 and 70,000) were all purchased from Aladdin Reagent Co., Ltd (Shanghai, China). Ascorbic acid, salicylic acid, sulfosalicylic acid, benzoic acid, sulfanilic acid, iron(III) chloride hexahydrate ($FeCl_3{\cdot}6H_2O$), dimethyl sulfoxide (DMSO) and absolute ethanol were from KeLong Chemical Reagent Co., Ltd. (Chengdu, China). Other reagents such as tris (hydroxymethyl) aminomethane (Tris), sodium hydroxide (NaOH), hydrochloric acid (HCl), sodium acetate trihydrate ($CH_3COONa{\cdot}3H_2O$) and acetic acid were all analytical grade. The ultrapure water used in this study was purified with an AK's laboratory pure water system from the Kangning Lab ultra pure Equipment Co., Ltd. (Chengdu, China).

2.2. Instrumentations

All the CE experiments were performed on an Agilent 7100 3D CE system (Agilent Technologies, Waldbronn, Germany) equipped with a diode array detector and an Agilent ChemStation software. Detection was performed at 200 nm for all of the analytes. Uncoated fused silica capillaries with 75 μm I.D., 375 μm O.D., 48.5 cm total length and 40.0 cm effective length were purchased from Ruifeng Chromatographic Devices Co., Ltd. (Yongnian, Hebei, China). UV–visible or fluorescence spectra were collected on an UV-2450 spectrophotometer and RF-5301 spectrofluorimeter (Shimadzu, Japan) to investigate the copolymerization of DA/PEI in solution. The morphology observation of the co-deposited columns were performed by FESEM (JEOL, JSM-7600F). The surface chemical components of the co-deposited coatings deposited on the surface of the capillary's raw material (quartz plate) were investigated using ATR-FT-IR spectroscopy (Nicolet iS50, Thermo Scientific Inc., USA).

2.3. Preparation of coating columns

The coating capillary columns were prepared via filling the capillary with a mixed aqueous solution of DA and PEI with different molecular weight, for a period of time. PEI, an amine-rich polymer, not only was used as the charge modifier, but also can react with DA through Michael addition or Schiff base reaction between amine and catechol groups [31]. Hence, the self-polymerization of DA and their copolymerization with PEI can be proceeded simultaneously. Afterwards, these co-deposited hybrid coatings were further incubated with Fe^{3+} to enhance their chemical stability [33]. The related co-deposition pathway is shown in Fig. 1. Briefly, new capillaries were preconditioned by flushing with methanol, 1 M HCl and 1 M