



ELSEVIER

Contents lists available at ScienceDirect

Talanta

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

Investigation on performance of zirconia and magnesia–zirconia stationary phases in hydrophilic interaction chromatography



Qing Wang^a, Jing Li^a, Xin Yang^a, Li Xu^a, Zhi-guo Shi^{b,*}, Lan-Ying Xu^c

^a Tongji School of Pharmacy, Huazhong University of Science and Technology, Wuhan 430030, China

^b Department of Chemistry, Wuhan University, Wuhan 430072, China

^c Hubei Key Laboratory of Economic Forest Germplasm Improvement and Resources, Comprehensive Utilization, Huanggang Normal University, Huangzhou, China

ARTICLE INFO

Article history:

Received 10 March 2014

Received in revised form

5 June 2014

Accepted 10 June 2014

Available online 19 June 2014

Keywords:

Zirconia

Magnesia–zirconia

Hydrophilic interaction chromatography

ABSTRACT

In the current study, zirconia (ZrO_2) and its composite, magnesia–zirconia ($MgO-ZrO_2$), were prepared as the hydrophilic interaction chromatographic (HILIC) stationary phases (SPs). Different experimental variables including water content, pH and buffer concentration in the mobile phase (MP) as well as column temperature were systematically studied to permit an in-depth understanding of the chromatographic properties of the mentioned SPs and to explore the retention mechanism further on. The results were compared with a native SiO_2 column. Adsorption was demonstrated as the main retention mechanism on the two ZrO_2 -based SPs. The transferring of the analytes from the MP to the ZrO_2 -based SPs was endothermic and high column temperature would facilitate the retention. In addition, the $MgO-ZrO_2$ SP exhibited superior resolution, column efficiency as well as stronger retention in comparison to the bare ZrO_2 SP, which demonstrated that the introduction of MgO could improve the structure and properties of the material. In conclusion, $MgO-ZrO_2$ was a promising material for HILIC applications.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

Zirconium dioxide (zirconia, ZrO_2), thanks to its remarkable thermal stability (can withstand high temperature up to 200 °C in chromatographic separation [1–3]), pH stability in the range of 1–14, mechanical stability as well as its particular surface chemical properties, has become one of the alternative metallic oxide-based chromatographic stationary phases (SPs) to silica (SiO_2) [4,5]. The surface of ZrO_2 is far more intricate than that of native SiO_2 , containing a number of distinct classes of interaction sites including Brønsted acid, Brønsted base and Lewis acid sites [6]. Zirconium ion (IV) is a strong Lewis acid that exhibits electropositivity and has an affinity for Lewis bases that donate lone pair electrons to form coordination compounds, which is called ligand-exchange interaction. Besides, ZrO_2 is an amphoteric metallic oxide, the hydroxyl groups of which can be protonated or deprotonated depending on the environmental pH, acting as either anion- or cation-exchanger respectively [7]. Owing to these special surface chemistry properties, ZrO_2 -based SPs exhibit distinct chromatographic properties compared to common chromatographic SPs like SiO_2 -based SPs. However, native ZrO_2 generally exhibits low

column efficiency due to the poor pore structure ('ink-bottle' shaped pores) and the chemically heterogeneous surface [8,9].

To improve the chromatographic performance of ZrO_2 -based SPs, hybrid ZrO_2 oxides, such as titania (TiO_2)– ZrO_2 [10], SiO_2 – ZrO_2 [11], ceria– ZrO_2 [12,13] and magnesia (MgO)– ZrO_2 [8,11,14], were developed. For example, $MgO-ZrO_2$ was demonstrated to possess enhanced specific surface area, improved specific pore volume and pore connectivity [8,14], resulting in higher column efficiency than the bare ZrO_2 SP [8,11].

Hydrophilic interaction chromatography (HILIC) was first named by Alpert [15], which was based on a polar SP and a relatively nonpolar binary eluent in which 3–40% [16] water acted as the stronger eluting component, and acetonitrile (ACN, a water-miscible organic eluting component) as the weaker one. Although there was still dispute about the retention mechanism in HILIC [17–19], an agreement that the retention mechanism was largely dependent on the SP, MP and the analytes was generally reached. HILIC has emerged as a complementary chromatographic mode to reversed-phase liquid chromatography (RPLC) especially for the separation of polar and hydrophilic analytes. However, the commercial HILIC SPs were almost based on silica or polymer matrices, and selection of HILIC SPs for application was random for lack of theory guidance. Therefore, it is important to prepare new SPs as alternatives to silica or polymer matrices, expand their applications, as well as investigate the retention mechanism in HILIC.

* Corresponding author. Tel.: +86 27 68752701; fax: +86 27 68754067.

E-mail address: shizg@whu.edu.cn (Z.-g. Shi).

As hydroxyl groups and zirconium ions (IV) exist on the surface, ZrO_2 is highly supposed to be an appropriate SP for HILIC. However, few reports were found about its application for HILIC. Kučera and co-workers [20,21] compared the HILIC retention behavior of a set of hydrophilic analytes on polybutadiene modified ZrO_2 , carbon-coated ZrO_2 , bare ZrO_2 and SiO_2 -based columns. In their study, adsorption and ligand-exchange interactions were found to be the primary retention mechanism on ZrO_2 -based columns, while partitioning was on the SiO_2 -based column. In addition, Randon et al. prepared a SiO_2 - ZrO_2 composite monolithic SP by generating ZrO_2 coating on the SiO_2 monolithic column [4]. The investigation of the retention mechanism revealed that partitioning was not the unique retention interaction on the SiO_2 - ZrO_2 monolithic column. Separation of three dimethylxanthine isomers which was difficult to be realized with RPLC was accomplished on the SiO_2 - ZrO_2 column in HILIC mode.

Considering the lack of systematic study on the retention mechanism of ZrO_2 SPs in HILIC, and there was no report on the application of hybrid ZrO_2 materials for HILIC, in this study, ZrO_2 and MgO - ZrO_2 were employed as HILIC SPs. Effects of several chromatographic parameters on retention of a set of test solutes were studied in detail to explore the retention mechanism of the two ZrO_2 -based SPs.

2. Experimental

2.1. Chemicals

Sulfamerazine, sulfamethazine, sulfathiazole, sulfadiazine, propranolol and procaine were obtained from Alfa Aesar (Tianjin, China). Isoniazide, adenine, phenytoin, berberine, thymine and quinine were purchased from Aladdin (Shanghai, China). Tetracaine and clonidine were bought from Tokyo Chemical Industry Co. Ltd. (Tokyo, Japan). ACN (HPLC grade), methanol (HPLC grade), acetic acid (HAc), ammonia, ammonium acetate (NH_4Ac), zirconium(IV) oxychloride octahydrate ($ZrOCl_2 \cdot 8H_2O$), magnesium chloride hexahydrate ($MgCl_2 \cdot 6H_2O$), ethanol, petroleum ether (60–90 °C) and isopropanol were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Polyoxyethylene sorbitan trioleate (Tween 85) and sorbitan monooleate (Span 80) were obtained from Shanghai General Chemical Reagent Factory (Shanghai, China). Ultrapure water was produced by a Heal Fore NW system (Shanghai, China).

2.2. Instrumentation

HPLC analysis was performed on a Dionex Ultimate 3000 chromatographic system (California, USA) which consisted of a degasser, two Ultimate 3000 pumps, an Ultimate 3000 automated injector, an Ultimate 3000 RS column compartment and a diode array detector. For data acquisition and analysis, the Chromeleon software was used. The Zeta potential was measured by a ZetaPALS high resolution Zeta potential analyzer (Brookhaven, USA).

2.3. Preparation of MgO - ZrO_2 and ZrO_2 columns

MgO - ZrO_2 microparticles were synthesized according to the procedures elaborated in the previous work [8,11]. Briefly, $ZrOCl_2 \cdot 8H_2O$ and $MgCl_2 \cdot 6H_2O$ were dissolved in pure water, and the mixture was then put into petroleum ether which contained Span 80 and Tween 85. The mixture was sheared and emulsified by a shear emulsifying mixer to form the emulsion. Ammonia gas was generated by heating ammonia aqueous solution and channeled into the emulsion for 2 h. White precipitation was obtained and separated by filtration, followed by washing with copious ethanol, petroleum ether and water. The microparticles were dried and sintered to burn off organic residues. Size classification was performed in a precipitation/decanting/resuspension order. Particles of 5 μm in diameter were collected, homogenized with isopropanol afterwards, and then pumped into the

stainless steel columns (150 mm \times 2.1 mm i.d., IDEX Health & Science LLC, MA, USA). ZrO_2 microparticles were synthesized as the same procedure of MgO - ZrO_2 without adding $MgCl_2 \cdot 6H_2O$. Scanning electron microscope (SEM) was carried out using a JSM-35CF instrument (JEOL, Tokyo, Japan). Fourier transform infrared spectroscopy (FT-IR) was performed on AVATAR 360 (Thermo, USA).

The bare SiO_2 column (150 mm \times 2.1 mm i.d., 5 μm) used for comparison was from Kromasil (Akzo-Nobel, Amsterdam, Netherlands).

2.4. Chromatographic evaluation

Stock solutions (1 mg/mL) of test solutes were prepared by dissolving the appropriate amount of each compound separately in methanol. These solutions were further diluted to get the working solutions (10 $\mu g/mL$) with the MP, prior to injection.

The following chromatographic conditions remained constant throughout the study: a flow rate of 0.2 mL/min, an injection volume of 5 μL . The column temperature was 25 °C except specified.

When NH_4Ac buffers were applied, the NH_4Ac concentration referred to the amount of NH_4Ac in the aqueous phase of the MP. Prescribed amount of NH_4Ac was dissolved in water to form the NH_4Ac solution and the required pH of it was adjusted by HAc or ammonia. And the pH value of the MP referred to the pH of aqueous phase, which was adjusted prior to mixing with ACN.

The void volume was calculated by dead time which was measured by injection of pure ACN. The retention time of the solutes was determined at least in triplicates all through the experiments. The measurement of retention was expressed as retention factor (k') for all the analytes.

2.5. Stability test

The stability test was performed with the MP of 97% ACN-3% of 50 mM NH_4Ac buffer at a pH of 6.0. k' and the theoretical plates of two probe analytes, i.e. sulfathiazole and sulfadiazine, were monitored to evaluate the stability of the two ZrO_2 -based columns.

3. Results and discussion

3.1. Characterization of synthesized MgO - ZrO_2 and ZrO_2 microparticles

SEM images of MgO - ZrO_2 and ZrO_2 microparticles are shown in Fig. 1. Both the two microparticles are spherical and in the size of $\sim 5 \mu m$. FT-IR spectra of ZrO_2 and MgO - ZrO_2 are depicted in Fig. 2. The absorption peak at around 3427 cm^{-1} should be ascribed to the vibration of -OH on the surface of MgO - ZrO_2 and ZrO_2 . In the spectrum of ZrO_2 , the absorption peaks appeared at 450.2, 514.2, 578.1 and 729.0 cm^{-1} corresponded to the stretching vibration of Zr-O. However, the strong characteristic peak of Mg-O at 500.1 cm^{-1} occurred in the spectrum of MgO - ZrO_2 , which masked the characteristic peaks of Zr-O. Based on the above analysis, ZrO_2 and MgO - ZrO_2 were successfully prepared.

3.2. Selectivity of the two ZrO_2 -based columns

In the preliminary experiment, a wide range of analytes containing acids (salicylic acid and its derivatives, benzoic acid, nicotinic acid and cyanuric acid), bases (sulfonamides, alkaloids, local anesthetics and nucleic acid bases) and neutrals (benzene, phenanthrene, naphthalene and biphenyl) were employed to investigate the selectivity of MgO - ZrO_2 and ZrO_2 SPs in HILIC mode. The acidic compounds showed asymmetric and deformed peaks on the ZrO_2 column; however, there was no signal on the

Download English Version:

<https://daneshyari.com/en/article/7680216>

Download Persian Version:

<https://daneshyari.com/article/7680216>

[Daneshyari.com](https://daneshyari.com)