Journal of Chromatography A, 1406 (2015) 331–336

Contents lists available at ScienceDirect

Journal of Chromatography A

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

Short communication

Liquid chromatographic method for toxic biogenic amines in foods using a chaotropic salt

Jian-Jun Zhong[a], Ningbo Liao[b], Tian Ding[a], Xingqian Ye[a], Dong-Hong Liu[a,*]

[a] Fuli Institute of Food Science, Zhejiang Key Laboratory for Agro-Food Processing, Zhejiang R & D Center for Food Technology and Equipment, Department of Food Science and Nutrition, Zhejiang University, Hangzhou 310058, China
[b] Department of Nutrition and Food Safety, Zhejiang Provincial Center for Disease Control and Prevention, Hangzhou 310006, China

ARTICLE INFO

Article history:
Received 30 April 2015
Received in revised form 6 June 2015
Accepted 14 June 2015
Available online 23 June 2015

Keywords:
Biogenic amine
Chaotropic salt
Food analysis
Ion-pair reagent
Retention behavior
Reversed-phase chromatography

ABSTRACT

Direct separation of biogenic amines by reversed-phase liquid chromatography (RPLC) is not an easy task because their basic and hydrophilic characteristics can lead to poor retention, column overloading, peak tailing, and hence low efficiency. Rather than routinely resorting to derivatization or using classical hydrophobic ion-pair reagents (IPR), this work proposes a new RPLC method making use of the chaotropic salt KPF_6 as inorganic additive to an acidic acetonitrilic eluent to remedy the difficulties. Amine retention, overload behavior, peak shape, and column efficiency were significantly improved. The use of excess KPF_6 led to a very slight decrease of amine retention. Depending on amine, the dependence of the logarithmic retention factor on the volume percent of acetonitrile could be reasonably linear or quite convex. Coupled with UV detection, the method was applied to trace analysis for six biogenic, aromatic or heterocyclic amines in three types of food after a sample cleanup, as necessary, by ion-pair extraction. The reliability of the whole analysis was demonstrated to be satisfactory. The proposed method outperforms existing methods in that it eliminates the need for long and cumbersome derivatization procedures without losing sensitivity; it also represents a good surrogate for classical ion-pair chromatography (IPC) because of the desirable hydrophilicity of chaotropic salts.

© 2015 Elsevier B.V. All rights reserved.

1. Introduction

Biogenic amines are bioactive organic bases mainly arising from undesirable microbial decarboxylase activity during the processing, storage or spoilage of food [1,2]. Based on structure, they are classified as aliphatic, aromatic or heterocyclic [3]. Many aromatic or heterocyclic amines possess well-documented toxicity when ingested in sufficient amounts, scombroid fish poisoning caused by histamine and the "cheese reaction" by tyramine and 2-phenylethylamine being the most notorious. Aliphatic amines are generally far less toxic, albeit can potentiate the toxicity of other amines [3,4]. Besides being a public health problem, biogenic amines have been proposed as microbiological quality indicators of certain foods: the Codex Alimentarius standards for fish provide histamine concentrations as indicators for decomposition, hygiene, and handling [3].

RP-HPLC with pre- or post-column derivatization has been the most common technique for simultaneously analyzing several biogenic amines [1]. Due to their lacking chromophores and fluorophores, derivatization (chromophore- or fluorophore-tagging) is indispensable to the direct photometric detection of aliphatic amines. However, this may not be the case for aromatic and heterocyclic amines whose UV detection is straightforward [5–7] provided the sensitivity and selectivity prove adequate for trace analysis. Besides the consideration of detection, the aim of a pre-column derivatization strategy to ease the separation should be underscored [1,8], because biogenic amines are highly polar bases that often exhibit poor behavior in reversed-phase chromatography (RPC). For the same reason, a post-column derivatization strategy is commonly combined with the adoption of ion-pair chromatography (IPC) using hydrophobic ion-pair reagents (IPR) like alkyl sulfonates [7,9].

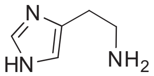
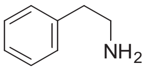
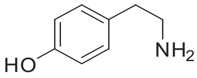
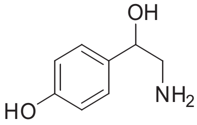
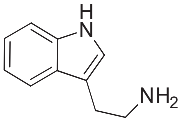
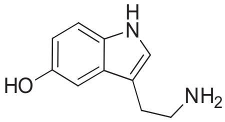
Chaotropic salts such as perchlorates, tetrafluoroborates, triflates, and hexafluorophosphates, are a new class of inorganic IPR for bases. Chaotropic salts mimic the role of hydrophobic IPR in improving the retention and peak shape of bases in RPC, while they are quite hydrophilic and easily dissolved in the mobile phase as opposed to hydrophobic IPR that tend to stick strongly to the

---

[*] Corresponding author at: Department of Food Science and Nutrition, Zhejiang University, Hangzhou 310058, China. Tel.: +86 571 88982169; fax: +86 571 88982154.
E-mail address: dhliu@zju.edu.cn (D.-H. Liu).

http://dx.doi.org/10.1016/j.chroma.2015.06.048
0021-9673/© 2015 Elsevier B.V. All rights reserved.

Table 1
Structures and chemical properties of biogenic amines.

Name	Structure	Chemical formula and molecular weight	pK_a	Log *P*
Histamine		$C_5H_9N_3 = 111.15$	5.9 (imidazole), 9.7 ($-NH_2$)	−0.70
2-Phenylethylamine		$C_8H_{11}N = 121.18$	9.83	1.41
Tyramine		$C_8H_{11}NO = 137.18$	9.5 (phenol), 10.8 ($-NH_2$)	(−0.14, 0.86)[a]
Octopamine		$C_8H_{11}NO_2 = 153.18$	8.81	−0.90
Tryptamine		$C_{10}H_{12}N_2 = 160.22$	10.2	1.55
Serotonin		$C_{10}H_{12}N_2O = 176.22$	10 ($-NH_2$), 10.7 (phenol)	0.21

[a] The two were predicted by ALOGPs and KOWWIN programs, respectively. All the other pK_a and Log *P* data were retrieved from the PHYSPROP database.

stationary phase, leading to slow column-equilibration and difficulty in recovering the initial column properties [10–13]. Nowadays, chaotropic salts are usually used in the analysis of active ingredients and impurities in pharmaceutical formulations [11,14–16]. In the meanwhile, attempts to understand the fundamentals behind the applications are still ongoing (see Ref. [17] for a review). Using $NaPF_6$ as mobile-phase additive, Flieger and Czajkowska-Żelazko investigated the chromatographic behavior of five neurotransmitter amines [18]. To the best of our knowledge, no approach has been proposed dealing with analyzing biogenic amines in foods, taking advantage of chaotropic salts.

As Erim [1] correctly concluded, the recent innovations in biogenic amine analysis by HPLC mostly concern extraction and derivatization procedures rather than separation science. The focus of this work, in contrast, is on developing a new RP-HPLC method making use of KPF_6 for analyzing six biogenic, aromatic or heterocyclic amines (Table 1) of toxicological relevance in food. For detection their natural chromophoric properties were straightforwardly exploited.

2. Experimental

2.1. Chemicals

Histamine, tyramine, and tryptamine were from Sigma–Aldrich (Shanghai, China); serotonin hydrochloride, 2-phenylethylamine, and octopamine hydrochloride were from Aladdin (Shanghai, China). Their purities were all over 98%. Bis (2-ethylhexyl) phosphate (BEHPA) 95% and KPF_6 99.98% were from Aladdin and HPLC-grade acetonitrile (ACN) from Scharlau (Barcelona, Spain). All other common chemicals (analytical grade) were from Sinopharm (Shanghai, China).

2.2. Chromatographic system and conditions

The HPLC system consisted of a Waters e2695 separation module, a 2489 TUV detector, and an Agilent Zorbax SB-C_8 column (150 mm × 4.6 mm I.D., 5-μm particles). The system void volume was 1.64 mL, determined by $NaNO_3$ injection with the test eluent 10 mM H_3PO_4–ACN 90:10. This value was used for all retention factor calculations. Peak tailing factors (TF) were calculated according to US Pharmacopeia recommendations.

The binary mobile phase consisted of (A) 50 mM KPF_6 in 10 mM H_3PO_4 (pH 2.2) and (B) ACN. The flow rate was 1.5 $mL\,min^{-1}$ and the column temperature maintained at 30 °C. The gradient was programmed as follows: 11–11–26–26% B at 0–5–5–11 min. A 5-min column-wash (with 100% ACN) and a 10-min equilibration were employed between successive runs. Typical injection volumes were 10 μL. The TUV detector was operated at dual-wavelength mode, 210 nm for quantitation and a different wavelength for coelution check. Baseline was autozeroed at 7.5 min.

2.3. Sample preparation

One vinegar and one *baijiu* (a Chinese liquor at 56% alcohol by volume) sample were purchased from a local store. A sample of chub mackerel, spoiled at room temperature for 2 days, was kindly provided by C. Wu.

The *baijiu* sample was diluted fivefold with deionized water and filtered before analysis.

Download English Version:

https://daneshyari.com/en/article/1199258

Download Persian Version:

https://daneshyari.com/article/1199258

Daneshyari.com