

RAPID CHROMATOGRAPHIC SEPARATION OF FOOD ADDITIVES ON THIN LAYERS OF AN INORGANIC ION-EXCHANGER

*K. Purghazi*¹, *V. Ghoulipour*¹, *S. Waqif–Husain*^{1,*}, and *A. Mirzaie*²

¹Department of Applied Chemistry, Faculty of Chemistry, University of Tarbiat Moallem, 49 Mofatteh Avenue, Tehran 15614, Iran

²Faculty of Food Science and Technology, Science and Research Branch, Islamic Azad University, P.O. Box 14515-775, Tehran, Iran

SUMMARY

The chromatographic behaviour of ascorbic acid, benzoic acid, butylated hydroxyanisole, butylated hydroxytoluene, butyraldehyde, butyric acid, cinnamaldehyde, citric acid, ethyl acrylate, ethyl benzoate, ethyl *p*-hydroxybenzoate, fumaric acid, lactic acid, lauric acid, maleic acid, methyl *p*-hydroxybenzoate, oleic acid, *p*-hydroxybenzoic acid, propionic acid, propyl gallate, propyl *p*-hydroxybenzoate, salicylic acid, sodium benzoate, and sorbic acid has been studied on thin layers of stannic silicate ion-exchanger with several aqueous, organic, and mixed mobile phases. Rapid separations of one food additive from many other food additives, and ternary and binary separations, are reported.

INTRODUCTION

Food additives are widely used to preserve the quality of the food, to achieve the uniformity needed for large-scale production, to enhance the flavour, or to improve the texture of a food product [1]. To monitor the use of additives in food many methods, for example spectrophotometry, voltammetry, and high-performance liquid chromatography, have been used during last two decades [2]. Because different types of additive are used for different purposes in one type of food product [3,4] selective separation is required before their quantitative analysis. Use of new synthetic inorganic ion-exchangers as stationary phases in thin-layer chromatography has transformed this technique into a powerful separation tool in chemical analysis [5]. We report here interesting results obtained from a study of the chromatographic behaviour of 24 food additives on thin layers

of a synthetic inorganic ion-exchanger, stannic silicate [6,7]. Rapid and selective methods have been developed for separation of one additive from the others. Binary and ternary separations of food additives have also been achieved.

EXPERIMENTAL

Chemicals and Reagents

All chemicals and reagents were of analytical grade from Merck, Fluka, and Riedel. The food additives used in this study were ascorbic acid (ASC), benzoic acid (BZA), butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), butyraldehyde (BUA), butyric acid (BUT), cinnamaldehyde (CIN), citric acid (CIA), ethyl acrylate (EAC), ethyl benzoate (EBE), ethyl *p*-hydroxybenzoate (EPHB), fumaric acid (FUM), lactic acid (LAC), lauric acid (LAU), maleic acid (MAL), methyl *p*-hydroxybenzoate (MPHB), oleic acid (OLE), *p*-hydroxybenzoic acid (PHBA), propionic acid (PRA), propyl gallate (PGA), propyl *p*-hydroxybenzoate (PPHB), salicylic acid (SAL), sodium benzoate (NBA), and sorbic acid (SOA).

Standard solutions (1 mg mL^{-1}) of the additives were prepared in ethanol. The solutions were protected from light and stored at 4°C .

Preparation of Ion-Exchange Plates

Aqueous solutions of stannic chloride (0.036 M) and sodium silicate (0.288 M with respect to Si) were mixed in the ratio 4:1 (v/v). The pH of the mixture was adjusted to 8.5 by addition of dilute aqueous ammonia solution and the mixture thoroughly stirred, during which a white gel was formed. The gel was left overnight then washed with demineralized water till the supernatant was free from ions. The supernatant was removed completely. A slurry, prepared by mixing 75 mL gel with 14 g silica gel G 60 powder, as binder, was used to coat seven $20 \text{ cm} \times 20 \text{ cm}$ glass plates with a $300\text{-}\mu\text{m}$ layer, by use of a Camag automatic TLC coater. The plates were dried in an oven at $60\text{--}70^\circ\text{C}$ for 2 h then stored at room temperature in a desiccator.

Chromatography

Sample solutions were applied to the plates as circular spots by means of disposable fine capillaries. The spots were dried completely and the plates were developed in ascending mode (without conditioning) in a Camag twin-trough chamber for $20 \text{ cm} \times 20 \text{ cm}$ plates. The development

distance from the origin was 12.5 cm. After development the plates were dried in a circulating air oven and the substances were detected by spraying with an aqueous solution of 1% KMnO_4 + 5% Na_2CO_3 (1:1, v/v). This had previously been found to be the most suitable detection reagent for the food additives; yellow to brown spots were obtained.

RESULTS AND DISCUSSION

The results listed in Tables I and II show that rapid and selective separation of the food additives can be achieved on stannic silicate ion-exchange plates. By using 1 M NH_4Br -acetone (3:7, v/v) as mobile phase (Table I) oleic acid, butylated hydroxytoluene, sorbic acid, sodium benzoate, and propyl gallate could be separated from many other additives in one development. Many rapid ternary and binary separations of the food additives were also achieved with simple aqueous systems as mobile phases (Table II). These separations are, in principle, based on selective adsorption of ionic and neutral species by the exchanger.

Table I

Separation of one food additive from others on thin layers of stannic silicate

Separation (R_T-R_L) [*]	Mobile phase	Interferences	Time (min)
OLE (0.42–0.62) from 16 additives	1.0 M NH_4Br -acetone, 3:7 (v/v)	LAU, BHT, CIA	44
MAL (0.70–0.84) from 16 additives	Chloroform-methanol, 4:6 (v/v)	LAC, FUM, PGA	33
ASC (0.90–0.98) from 12 additives	NH_4Br 0.5 M	LAC, LAU, FUM, SOA, SAL, MAL, CIA	14
BHT (0.48–0.70) from 16 additives	1.0 M NH_4Br -acetone, 3:7 (v/v)	LAU, EBE, BHA	44
SOA (0.48–0.80) from 14 additives	1.0 M NH_4Br - NH_3 , 3:7 (v/v)	FUM, CIN, EAC, PGA, SAL	14
NBA (0.00–0.00) from 14 additives	1.0 M NH_4Br -acetone, 3:7 (v/v)	PRA, BUT, LAU, EAC, CIA	44
PGA (0.48–0.80) from 14 additives	1.0 M NH_4Br - NH_3 , 3:7 (v/v)	FUM, CIN, EAC, SOA, SAL	14
BZA (0.24–0.41) from 13 additives	Chloroform (pure)	LAC, OLE, CIN, SOA, SAL, NBA	27
CIA (0.00–0.00) from 13 additives	Chloroform (pure)	LAC, MAL, CIN, PGA, SAL, ASC, NBA	27

^{*} $R_T = R_F$ of rear of spot, $R_L = R_F$ of leading front of spot

Table II

Ternary and binary separations achieved on stannic silicate plates

Mobile phase	Separation (R_T-R_L)*	Time (min)
Borate buffer (pH 9)	PPHB (0.45–0.65)–BZA (0.78–0.85)–PHBA (0.89–0.94)	15
	EPHB (0.67–0.77)–NBA (0.80–0.84)–PHBA (0.89–0.94)	
	PPHB (0.52–0.66)–SOA (0.70–0.78)–PHBA (0.87–0.92)	
	PPHB (0.47–0.65)–MPHB (0.77–0.82)–PHBA (0.90–0.95)	
	SAL (0.75–0.81)–PHBA (0.89–0.93)	
	PPHB (0.54–0.67)–NBA (0.80–0.85)	
KCl (1 M)	SOA (0.43–0.56)–MPHB (0.67–0.73)–PHBA (0.83–0.89)	18
	PPHB (0.36–0.46)–BZA (0.59–0.66)–PHBA (0.81–0.87)	
	EPHB (0.51–0.60)–MPHB (0.65–0.70)–PHBA (0.82–0.90)	
	PPHB (0.34–0.46)–SAL (0.60–0.74)–PHBA (0.81–0.87)	
Ammonium sulphate, 1%	BHT (0.00–0.00)–SOA (0.41–0.76)–PRA (0.80–0.91)	12
	BHT (0.00–0.00)–SOA (0.50–0.76)–BUT (0.80–0.91)	
	BHT (0.00–0.00)–SOA (0.45–0.75)–LAC (0.79–0.90)	
	BHT (0.00–0.00)–SOA (0.42–0.76)–MAL (0.80–0.94)	
	BHT (0.00–0.00)–SOA (0.55–0.75)–OLE (0.79–0.90)	
	BHT (0.00–0.00)–SOA (0.54–0.77)–BUA (0.81–0.90)	
	BHT (0.00–0.00)–SOA (0.53–0.77)–EBE (0.81–0.91)	
	BHT (0.00–0.00)–SOA (0.57–0.78)–EAC (0.82–0.93)	
	BHT (0.00–0.00)–SOA (0.49–0.76)–ASC (0.80–0.93)	
	BHT (0.00–0.00)–SOA (0.57–0.76)–CIA (0.80–0.91)	
NaH ₂ PO ₄ –NaOH buffer (pH 8)	PGA (0.00–0.00)–SOA (0.66–0.86)–PRA (0.92–0.98)	21
	BHA (0.00–0.00)–SAL (0.76–0.92)–PRA (0.96–1.00)	
	PGA (0.00–0.00)–SOA (0.58–0.86)–BZA (0.92–0.98)	
	PGA (0.00–0.00)–SAL (0.70–0.89)–FUM (0.96–1.00)	
	BHA (0.00–0.00)–SAL (0.76–0.91)–BUT (0.96–1.00)	
	PGA (0.00–0.00)–SOA (0.63–0.86)–LAC (0.92–0.98)	
	PGA (0.00–0.00)–SAL (0.73–0.91)–MAL (0.96–1.00)	
	BHA (0.00–0.00)–SAL (0.75–0.92)–BZA (0.98–1.00)	
	PGA (0.00–0.00)–SOA (0.58–0.84)–FUM (0.94–0.97)	
	BHT (0.00–0.00)–SAL (0.76–0.90)–LAU (0.96–1.00)	
	BHA (0.00–0.00)–SOA (0.58–0.76)–MAL (0.82–0.97)	
	PGA (0.00–0.00)–SAL (0.64–0.81)–LAU (0.96–1.00)	
	BHT (0.00–0.00)–SOA (0.62–0.77)–ASC (0.88–0.96)	
	BHA (0.00–0.00)–SAL (0.76–0.92)–BUT (0.95–1.00)	
Acetone–water, 4:6 (v/v)	CIA (0.00–0.40)–BUA (0.81–0.88)–EAC (0.96–1.00)	28
	BZA (0.00–0.00)–BUA (0.79–0.89)–EBE (0.96–1.00)	
	NBA (0.00–0.00)–BUA (0.78–0.87)–EAC (0.96–1.00)	
	PGA (0.00–0.00)–BUA (0.78–0.91)–SAL (0.96–1.00)	
	BZA (0.00–0.00)–BUA (0.78–0.86)–EAC (0.96–1.00)	
	NBA (0.00–0.00)–BUA (0.79–0.88)–EBE (0.96–1.00)	
	PGA (0.00–0.00)–BUA (0.76–0.85)–EAC (0.96–1.00)	
	CIA (0.00–0.44)–BUA (0.84–0.88)–EBE (0.96–1.00)	
	BHT (0.00–0.00)–BUA (0.84–0.87)–EAC (0.96–1.00)	
	PGA (0.00–0.00)–BUA (0.79–0.91)–EAC (0.96–1.00)	

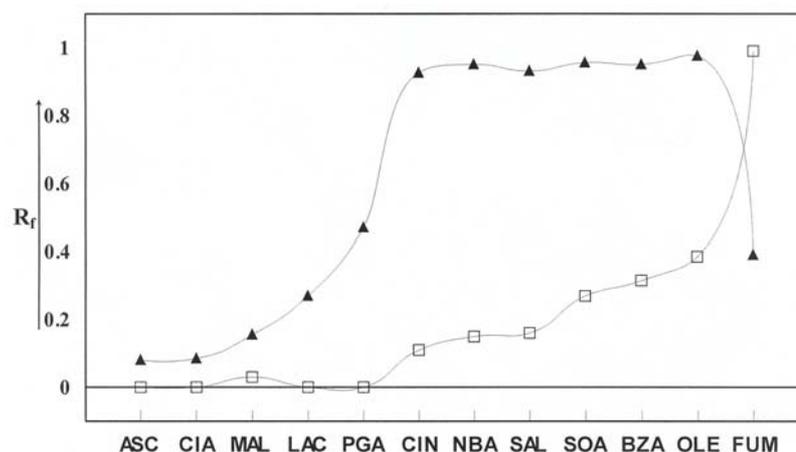
Table II (continued)

Ternary and binary separations achieved on stannic silicate plates

Mobile phase	Separation (R_T-R_L)*	Time (min)
Chloroform (pure)	MAL (0.00–0.05)–OLE (0.44–0.61)–PRA (0.96–1.00)	27
	CIA (0.00–0.00)–SOA (0.13–0.28)–FUM (0.93–0.97)	
	PGA (0.00–0.06)–OLE (0.38–0.52)–EBE (0.95–1.00)	
	ASC (0.00–0.00)–BZA (0.64–0.68)–BUA (0.95–1.00)	
	MAL (0.00–0.05)–OLE (0.42–0.54)–BUT (0.97–1.00)	
	ASC (0.00–0.00)–SOA (0.21–0.36)–LAU (0.93–0.97)	
	CIA (0.00–0.00)–OLE (0.34–0.48)–FUM (0.94–1.00)	
	PGA (0.00–0.07)–SOA (0.21–0.40)–BUT (0.93–0.97)	
	MAL (0.00–0.05)–OLE (0.42–0.56)–BHT (0.97–1.00)	
	CIA (0.00–0.00)–SOA (0.17–0.30)–PRA (0.91–1.00)	
	ASC (0.00–0.00)–OLE (0.35–0.44)–EAC (0.95–1.00)	
	MAL (0.00–0.07)–SOA (0.18–0.31)–PRA (0.93–0.97)	
	ASC (0.00–0.00)–OLE (0.42–0.58)–LAU (0.94–1.00)	
	PGA (0.00–0.08)–BZA (0.48–0.50)–BUA (0.93–0.97)	

* $R_T = R_F$ of rear of spot, $R_L = R_F$ of leading front of spot

Chromatography of the food additives with chloroform, chloroform–methanol, and chloroform–acetone mobile phases resulted in widely different R_F values (Figs 1 and 2), indicating the possibility of novel separations with non-aqueous mobile phases also.

**Fig. 1**

R_F values of the food additives developed with chloroform (squares) and with chloroform–acetone, 1:1 (v/v) (triangles)

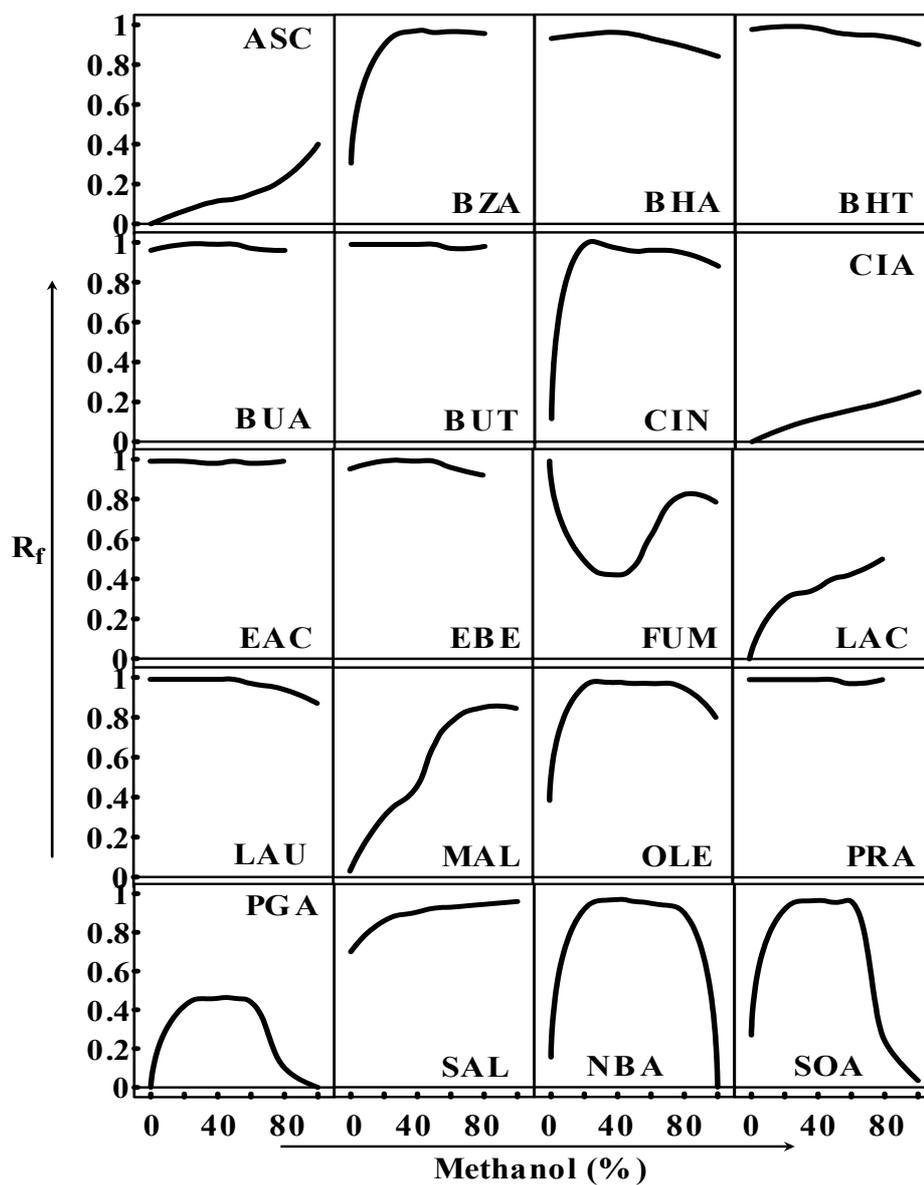


Fig. 2

Dependence of the R_F values of the food additives on the amount (%) of methanol in methanol–chloroform mobile phases

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