

Selective Liquid–Liquid Extraction of Natural Phenolic Compounds Using Amino Acid Ionic Liquids: A Case of α -Tocopherol and Methyl Linoleate Separation

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ABSTRACT: Amino acid ionic liquids (AAILs) with different amino acid anions were investigated in the selective separation of a typical natural phenolic product, α -tocoperol, from its mixture with methyl linoleate by liquid–liquid extraction. A large separation selectivity, suitable distribution coefficient, and adequate extraction capacity were achieved with the AAIL/N,N-dimethylformamide (DMF) mixture as extractant. The selectivity of α -tocopherol to methyl linoleate reached up to 29 when using [emim]Ala and [emim]Lys as the extractant diluted by DMF with mole ratio of AAIL to DMF 15:85, at least 9 times larger than that using DMF or common ILs as the extractant. The presence of diluents, DMF, can not only reduce the viscosity of IL phase, but could also lead to much larger distribution coefficients. Back extraction of α -tocopherol using hexane and reuse of AAIL were both tested. Solvatochromic and infrared spectra measurements were used to investigate the mechanism of α -tocopherol extraction with ILs. A close linear relationship can be drawn between the distribution coefficients of α -tocopherol and the hydrogen-bond basicity (β) of the extraction solvents, and also between the selectivites and the β values.

1. INTRODUCTION

Natural phenolic compounds are a class of important acidic bioactive products consisting of one or more hydroxyl groups (-OH) bonded directly to an aromatic hydrocarbon group, such as phenols, polyphenols, and flavonoids. Recently, interest has increased considerably to obtain naturally occurring phenolic compounds which are beneficial to human health and safer than the synthetic ones.¹ But it is difficult to isolate these natural acidic bioactive products from raw materials since they always exist in a mixture with various structurally related compounds.

A good example for the separation of natural phenolic compounds is the production of natural tocopherols. Tocopherols are the main components of Vitamin E, which is the most common and effective fat-soluble vitamin.² Tocopherols have many biological functions including anti-oxidant activity.³⁻⁵ They are found in varieties of vegetable oils, e.g. palm oil, rapeseed oil, and soybean oil, and coexist with large amount of free fatty acids. The methylated oil deodorizer distillate (ME-DOD), a byproduct from the vegetable oil refining process, is the main feedstock of natural tocopherols production.⁶ The key step to obtain tocopherols from ME-DOD is the separation of tocopherols and fatty acid methyl esters (FAMEs), which are the main components of ME-DOD.⁷ Because tocopherols have phenolic –OH groups that provide them with a relatively high hydrogen-bond acidity compared with FAMEs (their stuctures are shown in Figure 1), adsorption,^{8–10} ion exchange,¹¹ and solid phase extraction¹² are used for the separation based on the interaction between the hydroxyl group of tocopherols and the functional groups (e.g., -NH₂, -NH-, and -NHCO-) of adsorbents and ionexchangers. Distillation,¹³ supercritical fluid extraction,^{7,14,15} and organic solvent extraction¹⁶ are also reported in the separation of tocopherols. All of the above have some



Figure 1. Structures of α -tocopherol and methyl linoleate which are selected as the model of ME-DODs and the biphasic system consisting of hexane and IL/DMF mixture for extraction.

disadvantages: adsorption and ion exchange bear problems of low capacity and large consumption of solvents and adsorbents, distillation has a relatively low energy efficiency and low separation selectivity, and the selectivity for tocopherols obtained by supercritical fluid extraction and organic solvent extraction is low. Therefore, novel methods for effective separation of tocopherols from ME-DOD have been in great demand.

Recently, there is a growing interest in ionic liquids (ILs) due to their unique properties, such as negligible vapor pressure, high thermal and chemical stability, and the feasibility of structure and functional tunability.^{17,18} They have been applied in liquid–liquid extraction¹⁹ and elevated separation efficiency

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could often be obtained as compared to the traditional organic solvent extraction processes.^{20–25} The interactions between ILs and solutes can be finely adjusted by the change of the IL's cation or anion task-specificity, and ILs can readily form immiscible liquid–liquid biphasic systems with many solvents due to their high cohesive energy. So IL-mediated liquid–liquid extraction is potential for the separation of tocopherols.

Common ILs with PF_6^- , BF_4^- , Cl^- , etc. as anion have only weak or medium hydrogen-bond basicity (the hydrogen bond basicity of [bmim]Cl is 0.95), but have already shown remarkable effect on the extraction of acidic materials.^{16,26,27} In our previous work, tocopherol homologues (α -tocopherol and δ -tocopherol) were separated with a selectivity of 21.3 by using 1-butyl-3-methylimidazolium chloride as the extractant.28 Amino acid ionic liquids (AAILs) are a new type of functional ILs, which can be conveniently obtained by coupling an imidazolium cation with an amino acid anion.²⁹ Compared with common ILs, AAILs are reported to have relatively strong hydrogen-bond basicities (between 0.88 and 1.38). This is mainly because of the amino group $(-NH_2)$ on the anion which can form strong hydrogen-bonding interaction with acidic compounds. This unique property could make them effective in the extraction of phenolic compounds, including tocopherols. AAILs have been tested in solid phase extraction for tocopherols and showed good selectivity.¹² Liquid-liquid extraction, which proceeds between two different liquid phases, is a much more mature technology in large-scale industrial applications, with unique advantages, such as being easy to operate and scale up, not limited by the porous structure of solid medium during the transport of components, large throughput capacity, easy to carry out in a continuous mode, etc. Thus, using AAILs in the liquid-liquid extraction of tocopherols is more likely to be applied for industrial production.

Therefore, in this article, we describe the investigation of the selective separation of α -tocopherol and methyl linoleate by ILmediated liquid-liquid extraction. α -Tocopherol and methyl linoleate are selected as the model of ME-DOD (Figure 1). This model has been commonly used to evaluate the performance of new adsorbent or extractant for tocopherols separation.^{9,10} A new immiscible liquid–liquid biphasic system, consisting of AAIL/N,N-dimethylformamide (DMF) mixture and hexane, is involved in the separation. AAILs, with different basicity amino acid anions, are investigated along with several common ILs. Especially, Lys⁻ with double amino functional groups is employed. The presence of DMF as diluent of IL can not only decrease the viscosity of IL phase and the consumption of IL, but also increases the variety of the physicochemical properties through different IL/DMF compositions. The effect of the structure and concentration of IL, as well as other factors, the concentration of feedback, and the operation temperature on the extraction equilibrium were evaluated. AAILs investigated in our experiments have exhibited satisfactory efficiency in the separation of α -tocopherol and methyl linoleate by liquid-liquid extraction. Also solvatochromic and infrared spectra methods were used to study the physicochemical properties of extractant, and the extraction mechanism and property-extraction equilibrium relationship are discussed.

2. EXPRIMENTAL SECTION

2.1. Chemicals and Reagents. 1-Ethyl-3-methylimidazolium bromide ([emim]Br, 99%), 1-ethyl-3-methylimidazolium trifluoromethanesulfonate ([emim]TfO), 1-ethyl-3-methylimidazolium tetrafluoroborate ([emim]BF₄, 99%), and 1-ethyl-3-methylimidazolium hexafluorophosphate ([emim]PF₆, 99%) were obtained from Green Chemistry and Catalysis, LICP, CAS (China). The water mass fractions of [emim]Br, [emim]TfO, [emim]BF₄, and [emim]PF₆ were, respectively, 0.14%, 0.01%, 0.08%, and 0.01%, as determined by a Karl Fischer titrator. Glycine, lysine, alanine, serine, and proline were obtained from Aladdin (China). α -Tocopherol (99%) was purchased from Tokyo Kasei Kogyo Co., Ltd. (Japan). Methyl linoleate (99%) was obtained from Aldrich. Other chemicals (analytical grade) were all commercially obtained and used without further purification.

2.2. Preparation of Amino Acid Ionic Liquids. Ionic liquids with amino acids as anions were synthesized according to the procedure reported by Fukumoto et al. (Figure 2).³⁰



Figure 2. Preparation of amino acid ionic liquids.

First, the bromide anion of [emim]Br was exchanged with a hydroxyl anion by eluting the [emim]Br aqueous solution through strongly basic anion exchange resin. The elution of [emim]OH was tested by AgNO₃ aqueous solution to ensure the bromide anion was exchanged completely. Second, the [emim]OH aqueous solution was added dropwise into excessive amino acid aqueous solution and the neutralization occurred at room temperature under stirring for 24 h. The resulting aqueous solution was evaporated under vacuum at 60 °C to remove the water. Then the crude AAIL was dissolved in acetonitrile/methanol solution (the volume ratio of acetonitrile to methanol was 90:10) and the excessive amino acid was removed by filtration. The final product was dried under vacuum at 55 °C for 24-48 h. Ionic liquids, [emim]Ala, [emim]Gly, [emim]Lys, [emim]Pro, and [emim]Ser, were obtained through the above method. Their purities were confirmed by ¹H NMR and the water mass fractions of [emim]Ala, [emim]Gly, [emim]Lys, [emim]Pro, and [emim]-Ser were 0.732%, 0.621%, 0.868%, 0.847%, and 0.686%, respectively.

2.3. Extraction Equilibrium Experiments and Back Extraction Operation. The distribution of α -tocopherol and methyl linoleate in liquid—liquid biphasic system was determined as follows. Known amounts of α -tocopherol and methyl linoleate were dissolved in hexane to form the initial feedstock. Equal volumes of the hexane solution and an IL/ DMF mixture were mixed in an Erlenmeyer flask. The flask was shaken for 3 h in a thermostatic rotary shaker at 200 r/min, and then settled for 3 h at the same temperature. The shaking time was checked to be enough for extraction equilibrium. Samples

were taken by syringes without disturbing the phase boundary and diluted with the mixture of methanol and water (92/8, v/v)for the HPLC analysis. The extraction equilibrium experiments were repeated three times, and the relative uncertainties of distribution coefficient were within 5%.

For back extraction of α -tocopherol to hexane solution, an equal volume of hexane was added with 5 mL of the [emim]Ala/DMF solution (mole fraction of [emim]Ala 0.05) of α -tocopherol (15 mg/mL) and the two-phase mixture was shaken for 30 min and then settled for 30 min at the same temperature. Then the used hexane phase was removed and collected, and the remaining IL/DMF phase was extracted with the equal volume of fresh hexane again. The former steps were repeated four times, 20 mL of hexane was used, and the complete back-extraction was possible. Finally, the collected hexane phases were distilled under reduced pressure vacuum at 50 °C by a rotary evaporator. The samples taken from the concentrated hexane phase and the remaining IL/DMF phase were prepared for HPLC analysis.

To remove the DMF from the α -tocopherol product, water was added to the concentrated hexane phase and the mixture was shaken for 30 min. Volume of water five times that of the hexane phase was used. After settling for 30 min, the washed hexane phase was distillated under reduced pressure vacuum at 90 °C by a rotary evaporator. The dried product was diluted by methanol for GC-FID analysis.

2.4. HPLC Analysis. The HPLC system consisted of an autosampler, a SunFire C18 column (5 μ m, 4.6 × 150 mm), a Waters 1525 binary pump, and a Waters 2487 dual λ absorbance detector. The mobile phase was a mixture of methanol and water (92/8, v/v). The detection of α -tocopherol was performed at 292 nm and the detection of methyl linoleate was at 205 nm.

2.5. GC-FID Analysis. GC-FID method was used to detect the amount of DMF in the product. The system consisted of a gas chromatograph (GC750, FULI, China) equipped with a flame ionization detector (FID). Gas chromatographic separations were carried out on an Agilent HP-5 column (0.25 μ m, 30 m × 0.25 mm). The column temperature was programed from 50 °C (1 min) rising at 5 °C/min to 120 °C, rising at 30 °C/min to 250 °C, and rising at 10 °C/min to 300 °C (30 min). The linear velocity of carrier gas and the total flow rate were 36 cm/s and 50 mL/min, respectively. The injection port temperature was 310 °C and the temperature of FID was 300 °C.

2.6. Solvatochromic and Infrared Spectra Measurements. The Kamlet–Taft parameters of the extractant, IL/ DMF mixture, were determined with the probes N_iN -diethyl-4-nitroaniline and 4-nitroaniline for dipolarity and polarizability (π^*) and hydrogen-bond basicity (β).^{31,32}

Aliquots of probe-dichloromethane solution of a certain concentration were added to a small vessel, and then some ILcosolvent mixture was added after the full evaporation of dichloromethane. The obtained mixture was stirred thoroughly, and then was shifted to a quartz cuvette and placed in the sample cell of a Shimadzu UV-2550 UV-vis spectrophotometer. The maximum absorption wavelength of sample was recorded. The temperature of the cell was maintained by an external super circular water-bath at the same temperature as extraction equilibrium experiments. Scanning was repeated seven times for each measurement, and the average of each record was taken as the final value. The uncertainty of maximum absorption wavelength was ± 0.5 nm. The direct evidence of hydrogen-bonding interaction between the α -tocopherol and extractant was obtained using infrared spectra.³³ DMF solutions of a certain amount of α -tocopherol (20 mM) and varying amount of [emim]Ala were observed using a Nicolet 6700 FTIR spectrometer at a resolution of 4 cm⁻¹ and 32 scans per sample. The liquid cell included KBr windows with a Teflon spacer (optical path length of 0.5 mm).

3. RESULTS AND DISCUSSION

Because α -tocopherol has a phenolic –OH group that provides relatively higher hydrogen-bond acidity compared with methyl linoleate, several ionic liquids with different hydrogen-bond basicity were selected as extractant for the separation of α tocopherol and methyl linoleate. 1-Ethyl-3-methylimidazolium was chosen as the cation because it can easily couple with various anions to form functionalized ILs. Two classes of anions were evaluated in this work: common anions including Br, BF₄⁻, TfO⁻, and PF₆⁻, and amino acid anions including Ala⁻, Gly⁻, Lys⁻, Pro⁻, and Ser⁻. Because most of the studied ILs are highly viscous or even solid near room temperature (e.g., the viscosity of [emim]Gly is 450 cP at 25 °C³⁰ and [emim]Br is solid), in this study, DMF, a polar aprotic solvent, was chosen as the diluent for ILs to reduce the viscosity of IL phase and dissolve the solid ILs. The nonpolar solvent, hexane, was selected as the solvent for α -tocopherol and methyl linoleate to form a biphasic system with IL/DMF solution (Figure 1). Using hexane as a solvent could also limit the interfering solvent-solute hydrogen-bonding interaction.

3.1. Extractive Separation of α -Tocopherol and Methyl Linoleate Using Imidazolium ILs with Common Anions as Extractant. In this work, prior to the investigation on the extraction performance of AAILs, four 1-ethyl-3methylimidazolium ionic liquids with Br⁻, BF₄⁻, TfO⁻, and PF₆⁻ as anions were first selected as extractants for the separation of α -tocopherol and methyl linoleate. The distribution coefficient data of α -tocopherol and methyl linoleate in IL-DMF-hexane biphasic system at different IL/ DMF ratios have been determined and the results are presented in Table 1. The distribution coefficient of solute i (D_i) and the selectivity of solute i to solute j ($S_{i/j}$) are calculated as eqs 1 and 2:

$$D_i = C_i^{\ L} / C_i^{\ hex} \tag{1}$$

$$S_{i/j} = D_i / D_j \tag{2}$$

where C_i^{IL} and C_i^{hex} stand for the mass fractions of solute in the IL phase and in the hexane phase, respectively.

As the data show in Table 1, α -tocopherol exhibits higher values of distribution coefficient than methyl linoleate under all studied conditions. For example, while the mole ratio of [emim]Br to DMF is 5:95, the distribution coefficients of α -tocopherol and methyl linoleate are 0.45 and 0.151, respectively. Furthermore, the distribution coefficient of α -tocopherol and the selectivity of α -tocopherol to methyl linoleate obtained by different ILs follow the order [emim]PF₆ < [emim]BF₄ < [emim]TfO < [emim]Br under the same experimental conditions. This order is in agreement with the order of these ILs anions' hydrogen-bond basicity strengths.³⁴ This phenomenon is reasonable. As a result of the structural difference between α -tocopherol and linoleate, α -tocopherol has a relatively high hydrogen-bond acidity compared with methyl linoleate. Thus the α -tocopherol is more likely to be

Table 1. Distribution Data of α -Tocopherol and Methyl Linoleate in IL-DMF-Hexane Biphasic System^{*a*}

	ionic liquids							
mole ratio of IL to DMF	[emim] Br	[emim] TfO	[emim] BF ₄	[emim] PF ₆				
distribution coefficients of α -tocopherol								
5:95	0.45	0.35	0.28	0.18				
10:90	0.23	0.13	0.10	0.09				
15:85	0.08	0.05	0.04	0.03				
pure DMF	1.10							
distribution coefficients of methyl linoleate								
5:95	0.151	0.127	0.086	0.075				
10:90	0.065	0.060	0.04	0.046				
15:85	0.025	0.027	0.018	0.021				
pure DMF	0.36							
selectivity of α -tocopherol/methyl linoleate								
5:95	3.0	2.8	3.3	2.4				
10:90	3.3	2.2	2.5	2.0				
15:85	3.2	1.9	2.2	1.4				
pure DMF	3.1							

^{*a*}The initial concentrations of α -tocopherol and methyl linoleate in hexane (mg/mL) were 0.5 and 1.0, respectively. Volume ratio of IL phase and hexane phase was 1:1. Operation temperature was 30 °C.

extracted from hexane phase by IL via hydrogen-bonding interactions, and the value of distribution coefficient and selectivity will increase with the increase of IL's hydrogen-bond basicity.

As the data show in Table 1, the distribution coefficients of α -tocopherol and methyl linoleate obtained with IL/DMF mixtures as extractant are lower than those obtained with pure DMF. And as the concentration of IL in the IL/DMF mixture increases, the distribution coefficients decrease sharply. The selectivities of α -tocopherol to methyl linoleate using [emim]-Br/DMF mixtures as extractant are equivalent with the ones obtained with DMF and remain almost unchanged when the concentration of [emim]Br in the mixture increases. For the other three ILs, the selectivities of α -tocopherol to methyl linoleate are lower than those obtained with pure DMF, and a downward tendency is found against the increase of IL concentration. On one hand, α -tocopherol and methyl linoleate are both weak-polar compounds. Thus increased polarity of extractant goes against their distribution to extract phase. On the other hand, α -tocopherol is a hydrogen-bond donor that distinguishes it from methyl linoleate, so its distribution can be enhanced by the increase of hydrogen-bond basicity of extractant. All of the four kinds of studied IL have a larger polarity than DMF. The hydrogen-bond basicity of [emim]Br is close to that of DMF, but those of the other three ILs are lower than DMF. Thus, it is reasonable that the distribution coefficients of both α -tocopherol and methyl linoleate decrease with the increasing cocentration of each IL and the selectivity with [emim]Br does not reduce as per the other three ILs.

3.2. Extractive Separation of α -Tocopherol and Methyl Linoleate Using Imidazolium ILs with Amino Acid Anions. Because the ILs with amino acid anions exhibit stronger hydrogen-bond basicity³⁰ than many other ILs, five AAILs were tested for the separation of α -tocopherol and methyl linoleate, including [emim]Ala, [emim]Gly, [emim]Lys, [emim]Ser, and [emim]Pro (Figure 1). These ILs were selected due to the structural differences of their anions. Ala⁻ differs from Gly⁻ by a methyl side chain. Ser⁻ has an additional polar –OH group while Lys⁻ has two –NH₂ groups. Pro⁻ has a –NH group and a ring structure.

As the results show in Table 2, it is obvious that the distribution coefficients of α -tocopherol in the biphasic systems

Table 2. Distribution Data of α -Tocopherol and Methyl Linoleate in AAIL-DMF-Hexane Biphasic System

	amino acid ionic liquids							
mole ratio of IL to DMF	[emim] Ala	[emim] Gly	[emim] Pro	[emim] Lys	[emim] Ser			
distribution coefficients of α -tocopherol								
5:95	2.46	2.18	2.01	2.30	1.02			
10:9	2.21	1.57	1.86	1.72	0.70			
15:85	1.46	0.91	1.52	1.07	0.53			
pure IL	0.030		0.014 ^a	0.013 ^a	0.007 ^a			
pure DMF			1.10					
distribution coefficients of methyl linoleate								
5:95	0.17	0.16	0.18	0.16	0.15			
10:90	0.09	0.08	0.12	0.08	0.09			
15:85	0.05	0.04	0.07	0.04	0.044			
pure IL	_b		_b	_b	_b			
pure DMF	0.36							
selectivity of α -tocopherol/methyl linoleate								
5:95	14.3	14.5	11.2	14.7	6.9			
10:90	24.6	19.4	16.0	22.3	7.7			
15:85	29.2	24.4	22.1	29.6	12.0			
pure DMF			3.1					

^{*a*}The operation temperature was 60 °C. ^{*b*}The concentration of methyl linoleate was not detected in the IL phase.

containing AAILs as extractant are significantly higher than those containing common ILs or pure DMF, while the distribution coefficient of methyl linoleate does not differ much among different kinds of ILs. Therefore, compared to the common ILs studied previously, a much more significant difference between the distribution coefficients of α -tocopherol and methyl linoleate, i.e. a much larger selectivity, is obtained when using AAIL as extractant. Under the same condition, the distribution coefficient of α -tocopherol is much larger than that of methyl linoleate. For example, while the distribution coefficient of α -tocopherol is over 2.0 when using 5:95 AAIL/DMF mixture as extractant, that of methyl linoleate does not reach 0.2, so the majority of α -tocopherol could be extracted into the IL phase as the majority of methyl linoleate remains in the hexane phase. And then, it is reasonable to believe that α -tocopherol could be selectively separated from the model system in high purity by multistage extractions.

Compared with the data obtained by using pure DMF as extractant (Table 1), even a small amount of AAIL in the AAIL/DMF mixture could effectively improve the separation selectivity. When the mole fraction of [emim]Ala increases from 0 to 0.05, the distribution coefficient of α -tocopherol increases from 0.36 to 0.17 so the selectivity increases from 3 to 15. This result confirms that AAILs truly play a key role in the separation of α -tocopherol and methyl linoleate. It seems that the addition of AAILs could enhance both the polarity and the hydrogen-bond basicity of the IL/DMF mixture and so is expected to benefit the distribution of strong-polar or hydrogen-bond acidic compounds. Although methyl linoleate and α -tocopherol are both weak-polar compounds, α -tocopherol has peculiar hydrogen-bond acidity compared with

methyl linoleate. So, with the concentration of AAIL increasing in the extractant, the distribution coefficients of methyl linoleate decrease all the time while those of α -tocopherol increase at first before the succeeding decrease. It is important to note that although the distribution coefficient of both methyl linoleate and α -tocopherol decreases with the increase of AAIL concentration, the distribution coefficient of methyl linoleate declines more sharply than that of α -tocopherol, thus a higher selectivity of α -tocopherol to methyl linoleate could be obtained when using AAIL-DMF mixtures with relatively larger mole ratio of AAIL to DMF as extractant. For example, while the mole ratio of [emim]Ala to DMF increases from 5:95 to 15:85, the selectivity increases from 14.3 to 29.2.

The trend that distribution coefficients decrease with the increase of AAIL content in IL/DMF mixture but selectivity increases was further confirmed by the experimental results obtained when using pure AAIL as extractant. As shown in Table 2, under the same conditions with IL/DMF mixture mediated experiments, the distribution coefficient of α tocopherol obtained by pure [emim]Ala was only 0.03, while methyl linoleate could not be detected in the IL phase. Similarly, the distrbution coefficients of α -tocopherol obtained by [emim]Lys, [emim]Pro, and [emim]Ser were all below 0.015 while methyl linoleate was not detected in the IL phase, although the operation temparature was adapted to 60 °C due to their very high viscosity at 30 °C. However, despite the improved selectivity, pure AAILs are not feasible as extractant for the separation of α -tocopherol and methyl linoleate, as a result of the too-small distribution coefficients. Therefore, it is apparent that the AAIL/DMF mixtures are more effective and practical than pure AAIL as extractant, because the additional diluent DMF can not only lead to a much lower viscosity and more moderate operation conditions, but could also bring on elevated distribution coefficients of components with adequate separation selectivity. Moreover, the consumption of ILs can also be reduced by the use of mixtures.

In addition, as expected, the structure of amino acid anions has obvious impact on the extraction equilibrium (Table 2). The [emim]Lys exhibits a relatively large extractive selectivity among these AAILs. This is because the Lys⁻ has an additional $-NH_2$ group compared with the other amino acid anions. The distribution coefficient of α -tocopherol obtained by [emim]Ser is obviously smaller than the values from the other AAILs. Because Ser⁻ possesses an additional hydroxyl group compared to the other AA anions, the hydrogen-bond basicity of [emim]Ser is weakened and the polarity is enhanced, which does not favor the distribution of α -tocopherol.

3.3. Proposed Mechanism of α -Tocopherol Extraction with ILs. The microscopic solvent properties were studied for IL and DMF mixture. The physicochemical characteristics of the extractant can help to analyze the interaction between the α -tocopherol and IL and provide evidence for the mechanism of the selective separation for α -tocopherol and methyl linoleate using ILs.

Solvatochromic method has been widely used to determine the microscopic solvent properties of pure liquids or liquid mixtures.^{31,32} Among the various solvatochromic parameters determined by different probes, two widely used Kamlet—Taft parameters, the dipolar/polarity π^* and the hydrogen-bond basicity β , were studied for the extractant in this work. The π^* parameter was determined from the spectroscopic shift of *N*,*N*diethyl-4-nitroaniline using eq 3³⁵

$$\pi^* = 8.649 - 0.314\nu_{\text{DENA}} \tag{3}$$

where ν_{DENA} is the maximum absorption wavelength of *N*,*N*-diethyl-4-nitroaniline in the sample. The β parameter was determined from the spectroscopic shift of both *N*,*N*-diethyl-4-nitroaniline and 4-nitroaniline using eq 4³⁶

$$\beta = -0.357\nu_{NA} - 1.176\pi^* + 11.12 \tag{4}$$

where $\nu_{\rm NA}$ is the maximum absorption wavelength of 4-nitroaniline in the sample.

The hydrogen-bond basity (β) of the extractant, IL/DMF mixture (the mole ratio of IL to DMF was 5:95) in the α -tocopherol extraction, was measured using solvatochromic method. The distribution coefficients of α -tocopherol and the selectivities of α -tocopherol to methyl linoleate versus the β value of the extractant are plotted in Figure 3a and 3b. The



Figure 3. Relationship between β value of IL/DMF mixture and distribution data obtained by the extraction equilibrium experiments. The mole ratio of IL to DMF was 5:95. The operation temperature was 30 °C. (a) Distribution coefficients of α -tocopherol versus β value. (b) Selectivity of α -tocopherol versus β value.

conclusion can be drawn that the higher distribution coefficient of α -tocopherol and selectivity was obtained using the ionic liquid possessing the higher β value. Also the figures show a close linear relationship between distribution coefficients and β values, and also between selectivites and β values. This proves that the hydrogen-bond basity of ionic liquid, indicating the ability to form the hydrogen-bonding interaction with α tocopherol, is the key factor in the extraction. For amino acid ionic liquid, the $-NH_2$ group in the amino acid anion can form strong hydrogen-bond interaction with the -OH group so AAIL can selectively and effectively extract the α -tocopherol into the polar IL phase.

To obtain direct evidence of hydrogen-bonding interaction, we involved the IR measurement. DMF solutions were of a certain amount of α -tocopherol and varying amount of [emim]Ala, which were the same composition of the extractant phase. The IR spectra result was as Figure 4. Without



Figure 4. IR spectra for α -tocopherol. Solutions were prepared in DMF with α -tocopherol (15 mM) and varying levels of [emim]Ala, one of the AAILs used in the extraction. The lower frequency stretching peaks (3481 cm⁻¹) indicate the formation of hydrogenbonded α -tocopherol/IL complexes.

[emim]Ala in the solution, α -tocopherol had an absorbance peak at 3637 cm⁻¹, which related to the stretching of the nonhydrogen-bonded hydroxyl group.³³ And also there was a peak at 3471 cm⁻¹, which is assigned to the hydroxyl stretching of the hydrogen-bonded α -tocopherol/DMF complex.³³ This was because DMF has tertiary amine groups which can form weak hydrogen-bonding interaction with α -tocopherol. Adding [emim]Ala, the peak at the lower frequency increased with the amount of [emim]Ala increasing, which indicated that small amount of AAIL in the extractant can strengthen the hydrogenbonding interaction between the extractant and α -tocopherol impressively. And the peak at 3637 cm^{-1} decreased at the same time, indicating that the -OH group interacted with AAIL. And the interaction site of AAIL was most possible on the amino group of amino acid anions, because lone pair can be found in the nitrogen atom. These spectra indicated that [emim]Ala, and by analogy AAILs, can form a relatively strong hydrogenbonding interaction with α -tocopherol which improves the extraction efficiency.

So with the indirect solvatochromic and direct infrared measurement, we have a proposed mechanism of α -tocopherol extraction with ILs, which is based on the mechanism of hydrogen-bonding interaction between the solute and extractant.

3.4. Effect of Temperature on Extractive Separation. The distribution data of α -tocopherol and methyl linoleate in the [emim]Ala-DMF-hexane biphasic system at different temperatures are presented in Figure 5. It can be seen that the distribution is not notably influenced by the temperature. Only a slightly decreasing trend of distribution coefficient for both α -tocopherol and methyl linoleate is observed. The enthalpy change of the extraction processes, ΔH , can be



Figure 5. Distribution data of α -tocopherol and methyl linoleate in [emim]Ala-DMF-hexane at different temperatures. The initial concentration of α -tocopherol and methyl linoleate in hexane was, respectively, 0.5 and 1.0 mg/mL. Volume ratio of two phases is 1:1. The mole ratio of [emim]Ala to DMF was 5:95.

obtained from the slope of $\ln D$ vs 1/T using the following Van't Hoff equation:

$$\ln D = -\frac{\Delta H}{RT} + C \tag{5}$$

where *R* is the gas constant and *C* is a constant. For α -tocopherol, the process is enthalpically favored that the correlation coefficient was 0.95 and the calculated ΔH was -9.31 kJ/mol. But for methyl linoleate, the correlation coefficient of the linear regression was only 0.45. This was possibly because the distribution data of methyl linoleate were rather small and changed quite slightly when temperature changed. This showed we could use temperature to adapt the distribution of α -tocopherol while the distribution of methyl linoleate remained almost the same.

The change of temperature might play multiple effects on the distribution of the two materials, for example, the liquid—liquid phase equilibrium of IL-DMF-hexane ternary system and the solubilities of α -tocopherol and methyl linoleate in this biphasic system. And the hydrogen-bond basicity of IL often decreases with the increase of temperature.

3.5. Effect of the Concentration of α -Tocopherol and Methyl Linoleate on Extractive Separation. Experiments were also performed at different initial concentrations of α tocopherol and methyl linoleate in hexane with [emim]Ala/ DMF mixture as extractant. The equilibrium concentrations of α -tocopherol and methyl linoleate in the IL phase versus that in the hexane phase are plotted in Figure 6a. Both α -tocopherol and methyl linoleate do not reach saturation in the IL phase even though the concentrations of α -tocopherol and methyl linoleate are already quite high in the feedstock (close to the saturation of feedstock). As a result, the distribution coefficients in Figure 6b are almost constant even though the initial concentrations of α -tocopherol and methyl linoleate in the hexane phase are quite high at 100 and 43 mg/mL, respectively. So the concentrations of α -tocopherol and methyl linoleate had almost no effect on the distribution coefficients and extractive separation under the investigated condition. This result indicates that the extractant consisting of AAIL and DMF could have a large extraction capacity.

3.6. Back Extraction and Reusability of IL Phase. According to the equilibrium data in the previous experiments, using AAIL/DMF mixture as extractant, α -tocopherol can be



Figure 6. Distribution data of α -tocopherol and methyl linoleate in [emim]Ala-DMF-hexane biphasic system at different initial concentrations. Volume ratio of two phases was 1:1. The mole ratio of [emim]Ala to DMF was 5:95. The operation temperature was 30 °C. (a) Concentration in each phase at equilibrium. (b) Distribution coefficients versus concentration in hexane phase plotted according to (a).

separated from the mixture through multistage extraction. So to investigate the back extraction, the [emim]Ala/DMF solution (mole fraction of [emim]Ala is 0.05) of α -tocopherol (15 mg/ mL) was used. Hexane was used as the back extraction solvent. The [emim]Ala/DMF solution of α -tocopherol was extracted for several times using an equal volume of hexane each time. From the distribution coefficient of α -tocopherol obtained by the previous experiments, we can calculate the back extraction efficiency according to the following equations:

$$C_{i+1} = \frac{M_{IL} \cdot D \cdot C_i}{M_{IL} \cdot D + M_{Hex}}$$
(6)

$$E_{back} = 1 - \frac{M_{IL} \cdot C_i}{M_{\alpha - Toc}} \tag{7}$$

where C_i is the concentration of α -tocopherol in the [emim]Ala/DMF phase after *i* times of back extraction, *D* is the distribution coefficient of α -tocopherol obtained from the former experiments, M_{IL} is the mass of IL/DMF phase, M_{Hex} is the mass of hexane phase, and $M_{\alpha\text{-Toc}}$ is the initial mass of α -tocopherol in the IL/DMF phase. Using the distribution coefficient of α -tocopherol obtained in the former experiments (which is 2.46 in Table 2), after 5 times of back extraction at 30 °C, the calculated concentration of α -tocopherol according to

eq 6 in [emim]Ala/DMF was 4.53 mg/g and back extraction efficiency should be 71.16% calculated according eqs 6 and 7. Through calculation, back extraction efficiency would be over 92% after 10 times of back extraction. The experiment result, that the concentration of α -tocopherol in [emim]Ala/DMF was 4.22 mg/g and the experimental back extraction efficiency reached 73.13%, agreed with the calculation result well. So the calculation was reliable, and the complete back extraction of α tocopherol was possible with more than 10 times of back extraction. Besides, the hexane phase and the product α tocopherol were investigated by HPLC and no [emim]Ala was detected, which meant no IL lost from the IL/DMF phase into the hexane phase during the back extraction. The DMF dissolved in the hexane phase could be effectively removed by water washing. The amount of DMF in the final α -tocopherol product was less than 0.0132 mg/kg determined by GC-FID analysis, which was far below the maximum residue limits of extraction solvents in foodstuff (1 mg/kg) according to Official Journal of the European Union 2009. So the multistage back extraction using hexane can be an effective way to strip α tocopherol from the IL/DMF phase.

The remained IL/DMF after back extraction was tested for reuse. Four consecutive recycles were carried out with the results shown in Figure 7. During each recycle, a small amount



Figure 7. Recycle test of [emim]Ala/DMF extractant. The recycle test was operated at 30 °C. The mole fraction of [emim]Ala is 0.05 in the extractant.

of DMF was added because DMF slightly dissolved in hexane. It was observed that, with the successive recycling, the distribution coefficients of α -tocopherol and methyl linoleate, and the selectivity of α -tocopherol to methyl linoleate, dropped a little. The distribution coefficient of α -tocopherol dropped from the initial 2.46 to 2.37 which was less than 4% decrease. The selectivity of α -tocopherol to methyl linoleate dropped from the initial 14.7 to 14.4 which was less than 3% decrease. The recycled IL extractant still remained at high efficiency for the α -tocopherol extraction.

4. CONCLUSION

In this study, AAILs were used as novel media for the liquid– liquid extractive separation of a typical natural phenolic product, α -tocoperol, from its mixture with methyl linoleate in the presence of DMF as diluent. Large separation selectivity, suitable distribution coefficient, and adequate extraction capacity were achieved. A selectivity of α -tocopherol to methyl linoleate up to 29 was received when using [emim]Ala and

[emim]Lys as extractant diluted by DMF with a mole ratio of IL to DMF 15:85. Even though the mole ratio of IL to DMF was as small as 5:95, the selectivity could be about 14, which was almost five times larger than that obtained with pure DMF. The large selectivity is attributed to the specific hydrogenbonding interaction between the amino groups in AAILs with the hydroxyl group in α -tocopherol. The presence of DMF can not only reduce the viscosity of IL phase, but can also improve the distribution coefficients of components. When using the [emim]Ala/DMF mixture with a mole ratio of 5:95 as extractant, the distribution coefficients of α -tocopherol and methyl linoelate were at least 80 times larger than those obtained with pure [emim]Ala. Also in this work, the mechanism of α -tocopherol extraction using ILs was proved based on the hydrogen-bonding interaction between the solute and extrctant. The higher distribution coefficient of α tocopherol and selectivity was obtained using the ionic liquid possessing the higher β value. Also a close linear relationship was established between the distribution coefficients and the β values of extraction solvent, and also between selectivites and β values. The IR measurement offered direct evidence of the hydrogen-bonding interaction between α -tocopherol and AAIL. Overall, a promising method for the separation and concentration of tocopherols from methylated oil deodorizer distillate based on AAIL-mediated liquid-liquid extraction was revealed. Further work, including extraction with real methylated oil deodorizer distillate feedstock and regeneration of AAIL, is required.

It is a common problem that the bioactive phenolic compounds of interest often appear in a mixture with various structurally related compounds. So the present work is not only useful for the separation of α -tocopherol, but also worthy of being considered as a reference to the production of other bioactive phenolic compounds.

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Notes

The authors declare no competing financial interest.

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