Solvent Type Affects the Number, Distribution, and Relative Quantities of Volatile Compounds Found in Sweet Whey Powder

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ABSTRACT

This study compares the performance of diethyl ether, methylene chloride, methyl formate, and pentane in the analysis of volatile flavor components in sweet whey powder. Extracts were prepared from sweet whey powder using each solvent. Volatile components were isolated by solvent extraction followed by solvent-assisted flavor evaporation. Gas chromatography-mass spectroscopy, coelution with known standards, and retention indices were used to identify the volatile compounds. Sixty total compounds were either positively or tentatively identified across all 4 solvents, but the number, distribution between the molecular classes, and relative quantities detected depended on solvent type. The highest number, widest distribution, and greatest relative quantities were found using methylene chloride and methyl formate, whereas diethyl ether and especially pentane were noticeably less effective. Results are characterized using molecular-based characteristics of solvents and solutes including dipole moment, dielectric constant, Log P (octanol-water partition coefficient), polarizability, water solubility, and Lewis acidity/basicity. Polarity and acidity/basicity were the primary factors that determined solvent performance. This work establishes a molecular-level basis for the selection of solvents in the analysis of sweet whey powder flavors.

Key words: solvent, volatile, sweet whey powder

INTRODUCTION

The determination of flavor-active compounds in foods is an established area of research. Several different techniques exist to extract volatile compounds from a food matrix including extraction with organic solvents. Solvent choice is a critical factor in determining volatile compound recovery and efficiency of an extraction method. Three solvents that have been widely used in many food extractions include pentane (PN), methyl-

ene chloride (MC), and diethyl ether (DE). Diethyl ether has been one of the most frequently used solvents for extraction of volatile compounds over the last decade, but few studies provide a justification for its use. One solvent not typically used in volatile extractions is methyl formate (MF). The 4 solvents differ in extraction efficiencies due to differences in chemical and physical properties (Table 1). Diethyl ether is less effective at extracting polar compounds in a variety of foods primarily due to its highly nonpolar character. Methyl formate and MC would predictably extract more polar compounds due to their greater polar character, as expressed by their larger dipole moments and dielectric constants and lower Log P values. Because PN is relatively insoluble in water and very nonpolar, it would predictably extract greater numbers and relative quantities of increasingly nonpolar compounds. The compatibility between the molecular and physical properties of the solvent and volatile solute is critical in selecting an appropriate solvent.

Other factors affecting solvent selection include flammability, toxicity, cost, reactivity, and stability. Understanding the potential hazards (e.g., flammability, toxicity, and stability) during storage is critical because it requires more caution in its use and handling. Using the National Fire Protection Association (NFPA) rating system for flammable and combustible liquids, MC has the lowest flammability rating (NFPA = 1) whereas DE (NFPA = 4), PN (NFPA = 4), and MF (NFPA = 4) have the highest flammability rating, because of their high volatility and low ignition temperatures. Solvent toxicity is another determinant in solvent selection for extractions, especially in the case where sensory detection is utilized; for example, GC-olfactometry. Diethyl ether has an oral ($LD_{50(\text{rat})} = 1,215,000 \mu\text{g/kg}$) and inhalation ($LD_{50(\text{rat})} = 73,000,000 \mu\text{g/kg}$; 2 h) toxicity that is relatively similar to the oral ($LD_{50(\text{rat})} = 1,600,000 \mu\text{g/kg}$) and inhalation ($LD_{50(\text{rat})} = 88,000 \mu\text{g/kg}$; 30 min) toxicities of MC (National Research Council, 1995). However, inhalation of high concentrations of DE can cause sedation, unconsciousness, and respiratory paralysis; high concentrations of MC vapor ($>500,000 \mu\text{g/kg}$ for 8 h) can cause less detrimental effects such as lightheadedness, fatigue, and nausea (National Re-

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Table 1. Chemical and physical properties of commonly used solvents amenable to volatile compound extractions

Solvent	Boiling point ¹ (°C, 760 mmHg)	Dipole moment ²	Dielectric constant ³	Log P ¹	Polarizability ¹ ($\pm 0.5 \times 10^{-24} \text{ cm}^3$)	Water solubility ¹ (g/L, 25°C)	SB ⁴	SA ⁵
Diethyl ether	33.2	1.15	4.3	0.98	8.85	47.2	0.562	0.000
Methyl formate	32	1.77	8.5	-0.23	5.25	402	0.422	0.000
Methylene chloride	40	1.60	9.1	1.19	6.49	15.8	0.178	0.040
n-Pentane	35.2	0	1.8	3.41	9.99	0.031	0.073	0.000

¹Values obtained from Advanced Chemistry Development, Inc., 2006; Log P = log of octanol/water partition.

²Values obtained from Univ. Southern Maine (http://www.usm.maine.edu/~newton/Chy251_253/Lectures/Solvents/Solvents.html) and Buyong et al., 2000.

³Values obtained from ASI Instruments Inc., 2006.

⁴SB = Solvent basicity; values obtained from Catalan et al., 1996.

⁵SA = Solvent acidity; values obtained from Catalan and Diaz, 1997.

search Council, 1995). Methyl formate has an oral ($\text{LD}_{50(\text{rat})} = 1,500,000 \text{ } \mu\text{g/kg}$) toxicity similar to MC, and a lower inhalation toxicity ($\text{LD}_{50(\text{rat})} = 50,000,000 \text{ } \mu\text{g/kg}$; 30 min; Chemical Land 21, 2000). Pentane is moderately toxic (oral $\text{LD}_{50(\text{rat})} > 2,000,000 \text{ } \mu\text{g/kg}$; inhalation $\text{LC}_{50(\text{rat})} = 364 \text{ } \mu\text{g/kg}$; Chemical Land 21, 2000). Inhalation of PN may result in nausea, dizziness, or irritation of the mucous membrane of the nose and throat.

The only solvent that exhibits high hazards during storage is DE. Over time, it has a potential explosive hazard due to the formation of unstable peroxides promoted by exposure to oxygen, light, and strong oxidizing agents. Pentane, MC, and MF exhibit fewer hazards than DE during storage under normal conditions (National Research Council, 1995).

The level and spectrum of volatile compounds obtained after extraction and GC-MS analysis is expected to be highly dependent on the solvent selection. To date, few recent studies have considered the effects of solvent properties on the extraction of volatile compounds from foods. This study will compare the types and relative quantities of volatile compounds extracted from sweet whey powder (SWP) by DE, PN, MF, and MC as related to the molecular and physical properties of the solvents and solutes.

MATERIALS AND METHODS

SWP Composition

Commercial SWP (Cheddar cheese whey, US extra grade, Foremost Farms Inc., Baraboo, WI) aged for 4 yr under ambient conditions was selected for analysis. The pH was measured using a pH electrode (Accumet AB15, Fisher Scientific, Pittsburgh, PA) on a 6.5-g SWP sample in 100 mL of distilled water (Sithole et al., 2005). Free moisture content was determined by a standard oven-drying method (Marth, 1985). Fat content was determined by the Mojonnier method (method 989.05;

AOAC, 2000). Ash content was determined using a muffle furnace at 535°C (method 935.42; AOAC, 2000). The lactose content was determined using an enzymatic method (984.15; AOAC, 1995). Volhard's method (method 935.43; AOAC, 2000) was used to determine the salt content. The protein content was determined by the Kjeldahl method (methods 930.29, 991.20; AOAC, 2000). Sample color was determined by colorimeter (Colorquest 45/0, Hunterlab, Reston, VA). All analyses were done in triplicate. The average chemical characteristics ($\pm$ standard deviation) of the SWP samples were as follows: moisture = $4.72\% \pm 0.12$; protein = $13.2\% \pm 0.03$; fat = $1.04\% \pm 0.10$; lactose = $63.9\% \pm 0.4$; ash = $5.39\% \pm 0.05$; salt = $0.34\% \pm 0.05$; and pH = 5.58 ± 0.01 . Colorimeter results were $L^* = 77 \pm 0.15$, $a^* = 4.3 \pm 0.068$, and $b^* = 28 \pm 0.12$. Results were consistent with those reported by Banavara et al. (2003); thus, the SWP selected would be considered to be in legal compliance with respect to US compositional standards. The colorimeter values are reflective of whey that has undergone some browning, resulting in a possible reduction in lactose and protein levels.

Preparation of Extracts

Sweet whey powder (8.0 g) was extracted with 25 mL of each solvent in a tightly capped 40-mL vial for 1 h with mild, intermittent, manual shaking every 15 min. The 4 solvents used were DE (Sigma-Aldrich, Inc., St. Louis, MO), MC (Sigma-Aldrich, Inc.), MF (Acros Organics, Morris Plains, NJ), and PN (Acros Organics). At 1 h, the samples were centrifuged at $1,200 \times g$ for 2 min. The supernatant was pipetted from the vial and collected in a 100-mL glass vial with a Teflon-lined lid. The SWP was resuspended in an additional 25 mL of solvent and the extraction was repeated as above for a total of $3 \times 25 \text{ mL}$ of each solvent per SWP sample. The combined extracts ($\sim 75 \text{ mL}$) were distilled under

vacuum (709 mm Hg) at 50°C by solvent-assisted flavor evaporation to separate volatile compounds from non-volatile compounds (Engel et al., 1999). After distillation, external standards (1.8 ppm of 2-methyl-3-heptanone and 1.6 ppm of 2-methylpentanoic acid in 5 mL of methanol) were added to each extract to correct for injection errors. Samples were dried using approximately 1 g of anhydrous sodium sulfate (Fisher Scientific, Fair Lawn, NJ) before concentration to 0.5 mL under a stream of nitrogen. Duplicate extractions of an additional 2 separate SWP samples were completed with each solvent.

GC-MS

Volatile compound analysis was performed using a gas chromatograph (Agilent 6890N, Agilent Technologies, Inc., Palo Alto, CA) with a mass selective detector (Agilent 5973 MS, Agilent Technologies, Inc.). Data analysis was performed using the accompanying computer software (ChemStation Software Rev. D.02.00 SP1, Agilent Technologies, Inc.). Chromatographic separation of the volatile compounds was achieved using a fused silica capillary column (RTX-Wax, 30 m long $\times$ 0.25 mm i.d. $\times$ 0.5 μ m film thickness; Restek Corp., Bellefonte, PA). Helium was the carrier gas at a constant flow rate of 0.8 mL/min. The oven temperature was programmed from 40 to 220°C at a rate of 10°C/min with initial and final holding times of 5 and 10 min, respectively. Electron impact ionization was used with a voltage of 70 eV. An m/z scan range of 29 to 350 amu was collected at 4.44 scans/s. Samples (1 μ L) were injected in the splitless mode.

Identification of Volatile Compounds

Positive identification of odorants was made by 3 determinants: 1) mass spectra (NIST version 1.7 mass spectral database; Agilent Technologies, Inc.), 2) coelution with a known standard by injecting a series of alkanes (C_6 to C_{28}) into the GC using the conditions above, and 3) comparison of a calculated retention indices (**RI**) to a referenced Kovats index (**KI**; Kovats, 1958; Karagul-Yuceer et al., 2001, 2002; Acree and Arn, 2004; Mahajan et al., 2004; El-Sayed, 2005). Compound identity was considered tentative if only 1 or 2 of the 3 criteria matched. The degree of identification is reported in Table 2.

RESULTS

Sixty compounds were identified (29 positive and 31 tentative across all 4 solvents) and assigned into 1 of 10 classes of compounds based on their major common

moieties as follows: alcohols, ketones, carboxylic acids, aldehydes, amines, amides, esters, sulfur-containing compounds, aromatic compounds, and furanic compounds (Table 2). Solvent performance was compared based on the number, distribution, and relative quantities of compounds detected within each class. The highest number of compounds was detected using MC (42), followed by MF (37), DE (35), and PN (25).

The distribution of compounds was defined as the number of different compounds detected in each solvent extract within each class of compounds. A solvent was considered to exhibit distribution effectiveness if greater than 50% of the total compounds per class were detected at any concentration. For example, 5 of 6 ketones were detected using MC, 4 of 6 using MF, 3 of 6 using DE, and 2 of 6 using PN. Therefore, the solvents MC and MF were deemed effective for extraction of ketones, whereas DE and PN were not. Using this criterion, effective extraction distributions of the following flavor compound classes for each solvent are as follows: MC—ketones, carboxylic acids, aldehydes, amines, amides, sulfur-containing compounds and furanic compounds; MF—ketones, carboxylic acids, aldehydes, sulfur-containing compounds, and furanic compounds; DE—carboxylic acids, aldehydes, amides, and furanic compounds; and PN—esters and aromatic compounds. The widest distributions for all classes of compounds were found using MC and MF, whereas the distributions using DE and PN were the narrowest.

The relative percentage quantities extracted by the respective solvents were compared as follows. Each flavor compound quantity was expressed as a percentage of the average maximum standardized peak area for a given compound in each class (Figure 1 and Table 2). For example, for ketones, MC had the highest relative quantity of pantolactone of the 4 solvents (MC = 0.10, MF = 0.076, DE = 0.025, PN = 0.016) and was used as the divisor for all 4 relative quantities to determine the percentage of peak area (MC = 100%, MF = 76%, DE = 25%, PN = 16%). Using these latter percentages, if the percentage relative quantity of a given compound exceeded 50%, it was included in the total number of compounds recovered at high quantities for a particular class of compounds. For example, pantolactone was included as 1 of the 6 total ketones with relatively high quantities for MC and MF, but not for DE and PN. Finally, a solvent was considered effective in the extraction of relative quantities for a particular class of flavor compounds when greater than 50% of the total compounds in each class exceeded the 50% relative peak area. Using this criterion, effective detection of the following flavor compounds was found for each solvent: MC—ketones, carboxylic acids, amines, amides, and sulfur-containing compounds; MF—car-

Table 2. Alcohols, ketones, acids, aldehydes, amines, amides, esters, sulfur-containing compounds, aromatic compounds, and furanic compounds found in SWP using direct solvent extraction with diethyl ether, methyl formate, methylene chloride and pentane analyzed by GC-MS on a capillary polyethyleneglycol column (values in parentheses are standard deviations)

Compound	RI ¹	ID ²	Standardized relative peak area ³				Log P ⁴	BP ⁴ (°C at 760 mmHg)	WS ⁴ (g/L, 25°C)
			DE	MC	MF	PN			
Alcohols									
2-Pentanol	1,123	1, 2, 3	0	0	0	0.0066 (0.0100)	1.41	139	23.3
1-Butanol	1,147	1, 2, 3	0.44 (0.40)	0.033 (0.040)	0	0	0.88	118	66.3
2-Heptanol	1,286	1, 3	0	0.022 (0.012)	0	0	2.29	159	4.39
1-Octanol	1,551	1, 2, 3	0	0	0.29 (0.04)	0.062 (0.023)	3.00	195	0.95
Ketones									
2-Hexanone	1,083	1, 2	0	0	0.010 (0.116)	0	1.44	128	7.59
1-Hydroxy-2-propanone	1,298	1, 2	0.085 (0.028)	0.48 (0.52)	0.12 (0.01)	0	-0.78	146	553
Butyrolactone	1,631	1, 2, 3	0.020 (0.003)	0.032 (0.019)	0.039 (0.022)	0.020 (0.004)	-0.76	204	281
Pantolactone	2,029	1, 2, 3	0.025 (0.013)	0.10 (0.05)	0.076 (0.012)	0.016 (0.006)	-0.8	225	187
2-Pyrrolidinone	2,048	1, 2	0	0.024 (0.013)	0	0	-1.45	245	392
2,5-Pyrrolidinedione	2,458	1	0	0.017 (0.001)	0	0	-1.18	288	176
Acids									
Acetic acid	1,436	1, 2, 3	9.4 (3.9)	6.3 (4.9)	10.7 (0.3)	0.044 (0.010)	-0.29	117	1,000
Formic acid	1,485	1, 2	0.97 (0.42)	0.0083 (0.0028)	7.2 (0.7)	0	-0.54	101	1,000
Propionic acid	1,523	1, 2, 3	0.068 (0.021)	0.14 (0.07)	0.11 (0.01)	0.035 (0.008)	0.25	142	1,000
2-Methyl-propanoic acid	1,552	1, 2, 3	0.020 (0.007)	0.041 (0.021)	0	0	0.59	155	1,000
2,2-Dimethyl-propanoic acid	1,563	1, 2	0.0025 (0.0014)	0	0.041 (0.017)	0	0.94	166	185
Butanoic acid	1,609	1, 2, 3	0.14 (0.04)	0.35 (0.19)	0.25 (0.02)	0.21 (0.03)	0.78	164	578
3-Methyl-butanoic acid	1,652	1, 2, 3	0.036 (0.012)	0.10 (0.03)	0.061 (0.005)	0.065 (0.015)	1.12	175	210
Pentanoic acid	1,718	1, 2, 3	0.0067 (0.0024)	0.017 (0.008)	0.011 (0.003)	0.0075 (0.0017)	1.31	185	171
Hexanoic acid	1,828	1, 2, 3	0.098 (0.035)	0.27 (0.15)	0.21 (0.02)	0.10 (0.05)	1.84	205	55.3
Heptanoic acid	1,934	1, 2, 3	0.15 (0.08)	0.55 (0.31)	0.017 (0.004)	0	2.37	223	19
2-Ethyl-hexanoic acid	1,934	1, 2, 3	0.15 (0.08)	0.55 (0.31)	0	0	2.72	228	7.85
Octanoic acid	2,042	1, 2, 3	0.067 (0.028)	0.27 (0.21)	0.23 (0.07)	0.049 (0.029)	2.9	239	6.92
Nonanoic acid	2,146	1, 2, 3	0.015 (0.005)	0.047 (0.028)	0.045 (0.009)	0.013 (0.003)	3.43	255	2.68
(S)-2-Hydroxypropanoic acid	2,159	1, 2	3.0 (1.2)	0.64 (0.41)	2.6 (1.9)	0	-0.7	228	1,000
Decanoic acid	2,251	1, 2, 3	0.068 (0.044)	0.64 (0.33)	0.24 (0.11)	0.041 (0.031)	3.97	270	1.1
4-Oxo-pentanoic acid	2,317	1	0	0.084 (0.050)	0	0	-0.49	243	1,000
Benzoic acid	2,417	1, 2, 3	0	0.22 (0.14)	0.11 (0.05)	0	1.89	249	82.9
Dodecanoic acid	2,467	1, 2, 3	0.028 (0.013)	0.15 (0.05)	0.39 (0.12)	0	5.03	296	0.23
Phenylacetic acid	2,539	1, 2	0	0.048 (0.026)	0	0	1.5	266	65.7
Hexadecanoic acid	2,570	1, 2, 3	0	2.4 (1.2)	0.49 (0.03)	0.010 (0.003)	7.15	341	0.022
Tetradecanoic acid	2,634	1, 2, 3	0	0.033 (0.010)	1.0 (0.5)	0	6.09	320	0.062
Aldehydes									
Nonanal	1,394	1, 2, 3	0.0062 (0.0044)	0.0049 (0.0014)	0.022 (0.003)	0.016 (0.002)	3.56	191	0.084
1-Piperidinecarboxaldehyde	1,786	1, 2	0.0017 (0.0006)	0	0.027 (0.004)	0	0.24	223	20.5
1-Methyl-1H-pyrrole-2-carboxaldehyde	2,093	1	0	0.0069 (0.0031)	0	0	0.26	196	28.4
Amines									
2-Butanamine	1,333	1	0	0.0097 (0.0083)	0	0	0.75	62.9	1,000
1-Pyrrole-2-carboxaldehyde	2,023	1	0	0	0	0.0062 (0.0041)	0.64	219	12
5-Methyl-thiazole	2,132	1	0	0.31 (0.012)	0	0	0.9	142	7.86

Continued

Table 2 (Continued). Alcohols, ketones, acids, aldehydes, amines, amides, esters, sulfur-containing compounds, aromatic compounds, and furanic compounds found in SWP using direct solvent extraction with diethyl ether, methyl formate, methylene chloride and pentane analyzed by GC-MS on a capillary polyethyleneglycol column (values in parentheses are standard deviations)

Compound	RI ¹	ID ²	Standardized relative peak area ³				Log P ⁴	BP ⁴ (°C at 760 mmHg)	WS ⁴ (g/L, 25°C)
			DE	MC	MF	PN			
Amides									
2-Hydroxy-propanamide	1,333	1	0	0.0097 (0.0083)	0	0	−1.68	282	594
Acetamide	1,738	1, 2	0.0088 (0.0029)	0.17 (0.15)	0.046 (0.019)	0	−1.23	221	537
Formamide	1,772	1, 2	0.0023 (0.0021)	0.031 (0.016)	0.036 (0.015)	0	−1.51	211	898
Propanamide	1,792	1, 2	0.0019 (0.0007)	0	0	0	−0.7	222	240
Esters									
Butyl acetate	1,077	1, 2, 3	0.24 (0.18)	0	0	0	1.77	127	5.75
Ethyl nonanoate	1,532	1	0	0	0	0.10 (0.01)	4.43	220	0.045
Methyl decanoate	1,590	1, 3	0	0	0	0.0035 (0.0022)	4.43	224	0.045
Sulfur-containing compounds									
Dimethyl sulfoxide	1,569	1, 2	0	0.016 (0.011)	0.022 (0.009)	0	−1.35	189	337
Dimethyl sulfone	1,895	1, 2, 3	0.20 (0.09)	0.77 (0.38)	0.38 (0.04)	0.039 (0.017)	−1.19	241	188
Aromatic compounds									
Ethyl benzene	1,130	1, 2, 3	0	0	0	0.0097 (0.0044)	3.21	136	0.11
Propyl-benzene	1,211	1, 2, 3	0	0	0	0.014 (0.003)	3.74	161	0.031
1-Ethyl-2-methyl-benzene	1,229	1	0	0	0	0.069 (0.001)	3.67	164	0.036
Maltol	1,964	1, 2, 3	0.21 (0.14)	2.6 (2.1)	1.9 (0.48)	1.1 (0.5)	0.02	285	1,000
Phenol	1,993	1, 2, 3	0	0	0.12 (0.02)	0	1.48	182	102
2,3-Dihydro-3,5-dihydroxy-6-methyl-4H-pyran-4-one	2,267	1	0.032 (0.009)	0.20 (0.10)	0.11 (0.03)	0	−0.42	281	54.2
2,4-Bis(1,1-dimethylethyl)-phenol	2,270	1	0.037 (0.020)	0	0	0	4.86	265	0.092
Furanic compounds									
Tetrahydro-2-furanmethanol	1,501	1, 2	0	0	0.43 (0.05)	0	−0.39	177	202
2-Furanmethanol	1,646	1, 2, 3	0.033 (0.012)	0.049 (0.022)	0.060 (0.010)	0.040 (0.014)	0.2	170	71
2(5H)-furanone	1,758	1, 2	0.078 (0.093)	0.14 (0.05)	0.74 (0.38)	0	−0.84	204	319
2-Butyltetrahydro-furan	1,872	1	0	0	0	0.0094 (0.0030)	2.41	159	0.97
Tetrahydro-2-methyl-furan	2,215	1	0.026 (0.013)	0	2.1 (2.7)	0	0.82	79.9	36.9
5-(Hydroxymethyl)-furfural	2,474	1, 2	0.035 (0.030)	0.064 (0.031)	0.37 (0.08)	0	−0.45	292	47.6
Dihydro-4-hydroxy-2(3H)-furanone	2,577	1	0.13 (0.07)	0.83 (0.21)	0.54 (0.45)	0	−0.31	182	250

¹RI = Retention index.

²ID (identification of compounds): 1 = mass spectrophotometer (match quality >70%); 2 = authentic standard; 3 = reference Kovats index.

³Solvents: DE = diethyl ether; MC = methylene chloride; MF = methyl formate; PN = pentane.

⁴Values obtained from Advanced Chemistry Development, Inc., 2006. Log P = log of octanol/water partition; BP = boiling point; WS = water solubility.

Figure 1. Average percentage relative peak area of compound classes using different solvents.

boxylic acids, aldehydes, sulfur-containing compounds, and furanic compounds; DE—no classes; PN—esters. Methylene chloride and MF had the greatest relative quantities for all classes of compounds combined compared with PN and DE. In addition to esters, PN was effective at extracting relatively large quantities of hydrocarbon aromatics (e.g., ethyl benzene, propyl benzene, and 1-ethyl-2-methyl benzene). Relative quantities for alcohols were distributed approximately equally between each solvent.

A notable observation was that the boiling points of the 60 flavor compounds were all very high and most well over 100°C (Table 2) with only 2 exceptions: 2-butanamine (63°C) and tetrahydro-2-methyl-furan (80°C). Flavor compounds having lower boiling points certainly exist in SWP (e.g., diacetyl, boiling point = 88°C; Mahajan et al., 2004). However, because extraction is only one of the steps in the analysis of flavor

compounds, solvent evaporation by nitrogen sparging being another, there may be flavor compounds having boiling points less than 100°C present in the matrix, but these were not detected due to possible losses during sample preparation or possible coelution with the solvent peak.

DISCUSSION

To understand the pattern of flavor compounds found using each of the 4 solvents, the physical properties of the solvents and flavor compounds were considered. Three molecular descriptors were selected for volatile flavor compounds: Log P, polarizability, and water solubility (Figure 2 and Table 2). Five molecular descriptors were selected for solvents: dipole moment, dielectric constant, Log P, polarizability, and water solubility (Figure 2 and Table 1). Diethyl ether and MF are weak

Figure 2. Range for properties of compound classes and solvents. Log P = log of octanol/water partition; Log S = log solubility (g/L).

Lewis bases with MF being the weaker of the 2; MC and PN are neither Lewis bases nor acids. High polarity is indicated by a small Log P and polarizability value, large dipole moment and dielectric constant, and high water solubility. In general, MF is the most polar solvent followed by MC, DE, and then PN.

The physical and chemical properties of the solvents affected detection of the different classes of compounds with polarity having the largest effect on compound number, distribution, and relative quantities (Table 2). For example, polar compounds such as ketones, short-chain carboxylic acids, amines, amides, sulfur-containing compounds, and furanic compounds were best detected using either MF or MC, the most polar solvents (Figure 1). Methyl formate was better than MC at selecting for C1 to C4 acids, C11 to C16 acids, aldehydes, and furanic compounds, whereas MC was better at selecting for ketones, C5 to C10 acids, amines, amides, and sulfur-containing compounds. Because MF is a weak Lewis base, it was expected and demonstrated to be effective at extracting acids. Furthermore, MC is the only solvent that exhibits a very slight acidity because of its ability to accept electron pairs, allowing for the possibility of improved solvation of basic compounds such as amides and amines. This may explain the better performance of MC compared with MF in detecting amides and amines.

This research shows that amines were best detected using MC. No amines were detected using DE and only 1 in 4 amines was detected using PN. Mahajan et al. (2004) found minor concentrations of pyrazine and other amine compounds in fresh SWP (~14% of the total compounds) by extraction with 2:1 PN:DE. Perhaps the PN component of their solvent blend was responsible for the recovery of the amines. In another study, Ferretti and Flanagan (1971) used MC as the solvent to extract SWP and found that up to 38% of the total compounds reported were amines, further confirming the performance of MC as effective for extracting amines.

Considering that DE is one of the most commonly used solvents in the extraction of volatile flavors from foods, it was interesting to note its relatively poor effectiveness in volatile compound extraction compared with other less utilized solvents. Because DE possesses the most basic character of the solvents evaluated, it would have been expected to detect the highest relative quantities of acids. However, this was not observed and was probably due to its slightly less polar character (lower dielectric constant and dipole moment) compared with MC and MF, suggesting that polarity plays a larger role in recovery of acidic volatile compounds than does acidity/basicity.

Pentane showed the highest selectivity for extracting aromatics and long-chain esters that are both relatively nonpolar compounds. Pentane is the least polar of the 4 solvents based on dielectric constant, dipole moment, Log P, and water solubility. It also has no solvent acidity or basicity. Therefore, it was expected and observed that PN would be most effective at extracting nonpolar and long carbon chain compounds. It was not surprising that PN did not achieve high relative quantities in the other flavor compound classes that are generally more polar and more basic than the aromatics and long-chain esters.

The relative quantities of alcohols detected were approximately evenly distributed amongst all the solvents, which can be attributed to various physical and chemical properties. The alcohols detected are amphoteric and in general possess relatively high solvent basicity, moderate acidity, polarity, and polarizability, and low water solubility. Literature reports on general properties of alcohols can be applied to this research (Wypych, 2001): 1) polarizability increases with increasing n-carbon chain length, 2) basicity of n-alkanols increases with increasing chain length up to octanol, 3) acidity of n-alkanols decreases with increasing chain length up to nonanol, and 4) acidity of n-alkanols decreases dramatically with increasing alkylation at the atom that bears the hydroxyl group (e.g., from 1-butanol to 2-butanol). These properties exhibit competing forces between each solvent and may explain the even distribution of alcohols detected among the solvents.

The relative absence of flavor compounds having boiling points below 100°C can be attributed to a possible coelution of the compounds with the solvent peak and to the solvent evaporation step during sample preparation. During this latter step, the solvent is evaporated from approximately 75 to 0.5 mL—a 150-fold reduction. The loss of highly volatile flavor compounds during this step is governed by the relative volatility (**RV**) defined as the ratio of the vapor pressure of the solvent to that of the flavor compound. If $RV > 10$, then the loss of flavor compounds is small. Otherwise, the loss of volatile flavor compounds is significant and can be extreme. A Raleigh distillation calculation can be used to illustrate this point (King, 1980). Acetaldehyde, for instance (boiling point = 20°C), has an RV value of 0.5 to 0.7, and loss during this step is predicted to be greater than 99.9%. For furan (boiling point = 31°C), the RV is 0.7 to 1.0, and loss is predicted to be 99.6%. For formic acid (boiling point = 100.6°C), RV is 10 to 14, and loss is predicted to be 38%. In agreement with this calculation, formic acid was detected in this study. However, for compounds having a lower boiling point and RV, losses during solvent evaporation probably caused the lack of detection of these flavor compounds.

In future work, other solvents and combinations of solvents may provide additional information in this area of study. Because MC and MF performed best at extracting the highest number, widest distribution, and highest relative quantities of compounds, these solvents together might capture an even wider spectrum of compounds. Other solvents having a similar boiling point, but more acidic character, such as acetaldehyde or propionaldehyde, may also exhibit specific advantages.

CONCLUSIONS

The individual physical and chemical properties of the solvent and target flavor compound can only be used to a certain extent to predict extraction efficiency. This study demonstrates that no single molecular or physical property predominates as a predictor of the extraction performance against a particular class of compounds. However, it appears that solvent polarity has a great impact on flavor compound number, distribution, and relative quantities. Other factors such as food composition and matrix effects, pH adjustments, and solvent blending may be considered of interest in future work.

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