contains significantly more protein than the flesh (Hansen, 1970).

CONCLUSION

A standard-addition ion-selective electrode technique can be used to accurately and precisely determine bromide in the range of 1–100 mg/L in peach extract. With such a method we were able to detect bromide with a recovery slightly above 100% and with less variability than with a method based on a calibration curve. Bromide analysis in crude peach extract saves the time that was previously used in ashing, drying, and other preparatory processes. Use of BrSE costs about one-fifth that of neutron-activation analysis or X-ray fluorescence.

The detection limit in peach extract for a double-junction electrode coupled with the standard addition procedure appears to be below 0.2 mg/L. This is evidenced by a mean of 0.14 ± 0.06 mg/L for the peach extract blanks, where the mean is still more than double the standard deviation. However, the accuracy of this value was not determined. The concentration represented by the minimum potential unit of the potentiometer increases proportionately with C_x (eq 1). Therefore, one decimal place is lost from the accuracy of a concentration measurement in the standard-addition BrSE measurement of bromide as the concentration value increases 1 order of magnitude.

A BrSE can detect bromide in peaches resulting from MeBr fumigation. We also found that bromide does not significantly diffuse from skin to flesh under conditions that approximate those during storage or transport of peaches across the U.S.

ACKNOWLEDGMENT

We are most grateful to Drs. James Brownell and Barry Gump of California State University, Fresno, and Preston Hartsell, Elisabeth Gomez, and Lisa Riedle, Horticultural Crops Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, for their valuable assistance.

Registry No. Br^-, 24959-67-9; NaBr, 7647-15-6; MeBr, 74-83-9.

LITERATURE CITED

Abdalla, N. A.; Lear, B. *Commun. Soil Sci. Plant Anal.* **1975**, *6*, 489–494.

Banks, H. J.; Desmarchelier, J. M.; Elek, J. A. *Pestic. Sci.* **1976**, *7*, 595–603.

Basile, M.; Lamberti, f. *Meded. Fac. Landbouwwet. Rijksuniv. Gent* **1981**, *46*, 337–341.

Cammann, K. "Working with Ion-Selective Electrodes"; Springer-Verlag: New York, 1979; pp 121–161.

Conacher, H. B. S.; Chandra, R. K. *J. Assoc. Off. Anal. Chem.* **1974**, *57*, 801–803.

Durst, R. A. "Ion-Selective Electrodes", National Bureau of Standards Special Publication 314; U.S. Government Printing Office: Washington, DC, 1969; pp 375–414.

Getzendaner, M. E. *Cereal Sci. Today* **1961**, *6*, 168–270.

Gnanasunderam, C.; Triggs, C. M. New Zealand DSIR Entomology Division Report No. 4, Auckland, New Zealand, 1983, pp 1–30.

Hansen, E. "The Biochemistry of Fruits and Their Products"; Hulme, A. C., Ed.; Academic Press: New York, 1970; pp 147–158.

Koryta, J. "Ion-Selective Electrodes"; Cambridge University Press: New York, 1975; pp 68–86.

Pflaum, R. T.; Frohliger, J. O.; Berge, D. G. *Anal. Chem.* **1962**, *34*, 1812–1814.

Shrader, S. A.; Beshgetoor, A. W.; Stenger, V. A. *Ind. Eng. Chem.* **1942**, *14*, 1–4.

Winteringham, F. P. W.; Harrison, A.; Bridges, R. G.; Bridges, P. M. *J. Sci. Food Agric.* **1955**, *6*, 251–261.

Received for review July 2, 1984. Revised manuscript received May 13, 1985. Accepted July 23, 1985.

Isolation of Flavor Compounds in Model Systems by Countercurrent Continuous Dialysis

Yueh-Ing Chang* and Gary A. Reineccius

A 40-m tubing type continuous dialyzer was designed in this study. The dialysis rate of flavor compounds was substantially increased over that of a batch process. The effect on dialysis efficiency of varying flow rates of sample and solvent was studied. The ultimate purpose of flavor isolation could be accomplished by manipulating these two operating parameters. The effects of solvent composition, polarity, molecular weight, and acidity of flavor components were also investigated. The more polar and smaller flavor compounds exhibited greater diffusivities. The addition of 2% (v/v) ammonium hydroxide or 1% (v/v) water in diethyl ether was found to be beneficial in isolation of basic flavor components.

INTRODUCTION

The selection of a flavor-isolation technique depends on the nature of the starting material, the flavor constituents of interest, the precision required, time available, and cost. The general methods of flavor isolation have been reviewed by several workers (Jennings and Rapp, 1983; Schreier, 1984; Reineccius, 1984; Reineccius and Anandaraman, 1984). These methods take advantage of the differences in volatilities and solubilities of the flavor compounds vs. the other constituents. For fatty foods, the flavor-isolation process is confronted with problems that are not present in fat-free systems. For example, the vapor pressure of flavor constituents was found to be up to 500 times less in the presence of fats than in totally aqueous systems (Buttery, 1973). Fats may undergo hydrolysis in the presence of water such as in steam distillation (Honkanen and Karvonen, 1966). Fats as well as flavors are extracted if solvent extraction is attempted, since they are mutually soluble (Arnold and Barnhart, 1972). The fats present in a flavor extract will stay in the injection port of a gas chromatograph or column and decompose by heat, thereby interfering with the analysis of the flavor profile (Chang et al., 1977).

To obtain fat-free flavor isolates and make solvent extractions applicable to fatty foods, Benkler and Reineccius (1979, 1980) explored a dialysis method to separate flavor

Department of Food Science and Nutrition, University of Minnesota, St. Paul, Minnesota 55108.

0021-8561/85/1433-1168$01.50/0

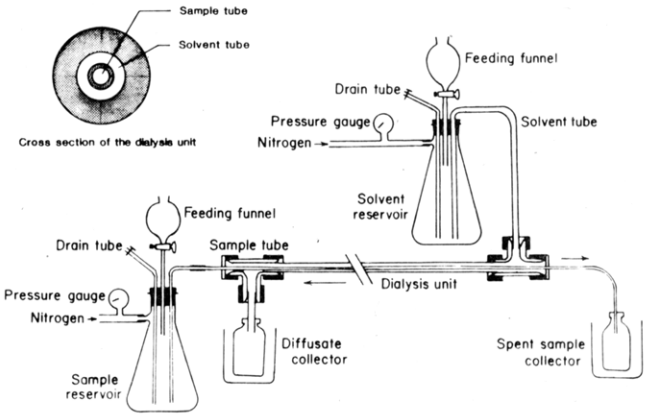

Figure 1. Schematic diagram of countercurrent dialysis system.

components from fats after solvent extraction of fatty food samples. This technique mainly utilizes the difference in molecular size between components. They used a batch dialysis cell. Thie results showed that many of the flavor compounds could be dialyzed out while fats were retained. However, the following problems still remained unresolved: (1) Since a batch system was used, equilibration required at least 24–72 h. (2) Theoretical maximum recoveries could reach only 50%. (3) Differential diffusion rates of flavor compounds across the membrane might lead to incomplete isolation. (4) Basic compounds failed to dialyze through the membrane. (5) Artifact formation was observed due to the membrane catalysis.

In order to improve dialysis as a separation technique, the present study had the following objectives: (1) to explore the possibilities of a continuous dialysis system; (2) to study the operating factors affecting dialysis efficiency; (3) to study the effects of chemical properties and molecular size of flavor compounds on dialysis efficiency; (4) to attempt to dialyze basic components using an acidic membrane.

MATERIALS AND METHODS

Dialysis Tubing. A 40-m perfluorosulfonic acid thin-wall tubing (Nafion 811X, E. I. du Pont de Nemours and Co.) of 1100 equiv wt was used. The equivalent weight is defined as the weight of the polymer that, when in the acid form, will neutralize 1 equiv of base. This dialysis tubing was stable in organic solvents (Grot, 1972) and had a nominal outside diameter of 0.875 mm and inside diameter of 0.625 mm when supplied.

Continuous Dialysis Module. A schematic diagram of the module is shown in Figure 1. It consisted of five basic parts: dialysis unit, sample reservoir, solvent reservoir, spent sample collector, diffusate collector. Spent sample represents the sample depleted of the diffused components, and diffusate refers to the solvent containing the diffused components after the dialysis process. Ice water baths were used for the dialysis unit and the two collectors to prevent vapor block or evaporative loss of solvent and volatiles.

The dialysis unit was formed by two 40-m coaxial tubings (Figure 1). The inner tubing was the dialysis tubing described above, and the outer tubing was a Teflon tube (i.d. = 1.588 mm, o.d. = 3.175 mm, Cole-Parmer Instrument Co.). Due to the fragile nature of the dry membrane, a special technique was adopted to assemble the dialysis unit. A monofilament fishing line (10 lb, Berkeley Co.) was first passed through the Teflon tube with facility by applying vacuum (5 mmHg) to one end of the tube. The dialysis tubing was then attached to the fishing line and gently pulled through the Teflon tube. Ancillary securing nuts, ferrules, and tee unions were subsequently used to make the unit leakproof, leaving the two ends of both tubings free. The sample to be dialyzed was passed through the inner tube (sample tube), and solvent was passed through the channel (solvent tube) between the two tubings, in all the experiments conducted.

A 1-L filter flask fitted with a three-holed stopper was used as the sample reservoir. Through the three holes were fitted respectively a drain tube (to drain excess sample and to clean the reservoir in place), the feeding funnel, and the sample tube. To the side arm of the flask, a short length of Tygon tubing and a pressure gauge were connected in series in order to apply and monitor the static pressure head of nitrogen gas. Flows were controlled by the constant head pressure of nitrogen in the reservoirs. An identical setup was used as the solvent reservoir. Two 250-mL glass bottles were used as the spent sample and diffusate collectors, in which a short length of the sample or solvent tube at the exit was immersed.

Countercurrent Dialysis. Reservoirs and collectors were attached to the dialysis unit in such a way as to achieve countercurrent flows of sample and solvent as shown in Figure 1. A general scheme for the countercurrent dialysis experiment was as follows: Prior to dialyzing the sample, the system was cleaned up and conditioned by passing 50 mL of selected solvent through both tubes under the conditions to be employed for the sample dialysis. Sample and solvent were then added in their respective reservoirs. Flows were initiated and regulated. All the experiments were conducted at a fixed volume (250 mL) of spent sample collected unless otherwise specified. The diffusate volumes varied from 140 to 2100 mL, depending upon the flow rates employed. Spent sample and diffusate were collected and used for further qualitative and quantitative analyses.

Preparation of Model Flavor Systems. Concentration of a flavor compound in each sample was 1 mg/mL, unless otherwise stated. All the compounds used were obtained from Sigma Co. (>99.9% purity). All the solvents were obtained from Fisher Scientific Co. (analytical grade).

All the experiments from hereon were done in duplicates. The results are averaged values.

Flow Rate Effect. Preliminary experiments indicated that the relative flow rates of sample and solvent affected dialysis efficiency. Four sets of experiments using different solvent flow rates were conducted. These selected solvent flow rates were 1.2, 1.7, 3.4, and 5.5 mL/min. For each solvent flow rate, three to five different sample flow rates (range 0.2–3.6 mL/min) were used when experimental conditions permitted.

The compounds used in this study were butanol, 2-octanone, tetradecane, 2-decanone, nonanol, decanol, and octadecane. Anhydrous diethyl ether containing 1% methanol (v/v) was used as the solvent.

Three measurements relating to dialysis efficiency were examined. They were concentration, dialysis rate, and fraction recovered of the test compounds in the diffusate. Concentrations of flavor compounds in the diffusate were measured via gas chromatography by the internal standard method. *n*-Nonyl acetate was used as the internal standard. The other two measurements were calculated by using the following formulas:

$$R_D(Y) = [C_D(Y)]Q_S$$

$$F_D(Y) = R_D(Y)/R_F(Y) = R_D(Y)/[[C_F(Y)]Q_F]$$

where $C_D(Y)$ = concentration of a compound (Y) in the diffusate (mg/mL), $R_D(Y)$ = dialysis rate of a compound (Y) across the membrane (mg/min), $F_D(Y)$ = fraction re-

covered of a compound (Y) in the diffusate, $C_F(Y)$ = feeding concentration of a compound (Y) (mg/mL), $R_F(Y)$ = feeding rate of a compound (Y) (mg/min), Q_S = volumetric flow rate of solvent (mL/min), and Q_F = volumetric flow rate of sample (mL/min).

Estimation of Diffusivity. Dialysis rate (R_D) has been found to be approximately proportional to the logarithmic mean concentration difference between sample and solvent fluids (ΔC_{LM}) for a given compound (Michaels, 1966). This relationship is shown by

$$R_D(Y) = 60[K(Y)]A[\Delta C_{LM}(Y)]$$

where $R_D(Y)$ = dialysis rate of a compound (Y) across the membrane (mg/min), $K(Y)$ = diffusivity of a compound (Y) across the membrane (cm/s), A = surface area of membrane (cm^2), $\Delta C_{LM}(Y)$ = logarithmic mean concentration difference between sample and solvent fluids for a given compound (Y) (mg/mL) $[=(\Delta C_1 - \Delta C_2)/[\ln(\Delta C_1/\Delta C_2)]]$, $\Delta C_1 = C_{\text{sample feed}} - C_{\text{diffusate}} = C_F - C_D$ (mg/mL), and $\Delta C_2 = C_{\text{sample feed}} - C_{\text{solvent feed}} = C_{SS}$ (mg/mL).

To utilize this relationship, ΔC_{LM} was calculated according to the formula given above for each compound in 17 dialysis experiments in the flow rate effect study. The slope of dialysis rate (R_D) vs. the logarithmic mean concentration difference (ΔC_{LM}) was calculated. Diffusivity [$K(Y)$] of each compound was then estimated from the slope since surface area of membrane (A) was known (ca. 1100 cm^2).

Molecular Weight Effect. A homologous series of alcohols (ethanol, propanol, butanol, pentanol, hexanol, heptanol, octanol, nonanol, decanol) and fatty acids (acetic acid, propionic acid, *n*-butyric acid, *n*-valeric acid, *n*-caproic acid, *n*-caprylic acid, *n*-capric acid) were used. The solvent used was anhydrous diethyl ether containing 1% methanol (v/v). Sample and solvent flow rates were 1.1 and 2.2 mL/min, respectively. The concentration of each alcohol and fatty acid in the diffusate was determined by using 2-octanone and *n*-undecane as internal standards, respectively.

Solvent Composition Effect. Three solvent compositions were studied: anhydrous diethyl ether, 1% methanol/anhydrous diethyl ether (v/v), and 5% methanol/anhydrous diethyl ether (v/v). The test compounds used in this study were hexanal, butanol, 2-octanone, tetradecane, propionic acid, nonanol, benzyl acetate, and octadecane. Sample and solvent flow rates were 1.6 and 2.0 mL/min, respectively. Dialysis was done for 1 h, and then the concentrations of compounds in the diffusate were measured with isoamyl acetate as an internal standard.

Basic Compound Dialysis. As noted by Benkler and Reineccius (1979), basic compounds were adsorbed onto the dialysis membrane and thereby not recovered from the system. A series of experiments were conducted in order to circumvent this problem. Since some of the experimental treatments could permanently damage the dialysis membrane, these experiments were conducted by using the batch system (Benkler and Reineccius, 1979).

Part 1. Effect of Ammonium Hydroxide Addition on the Dialysis of Basic Compounds. Four different solvent compositions were studied: anhydrous diethyl ether (control); 1%, 2%, and 5% ammonium hydroxide/anhydrous diethyl ether (v/v). The sample was composed of three basic compounds: pyrazine, 2-methylpyrazine, and 2,4,6-trimethylpyridine at a concentration of 2 mg/mL of each compound. The procedure of Benkler and Reineccius (1979) for a batch dialyzer was followed. *n*-Decane was incorporated as an internal standard. Diffusate and spent samples were analyzed after 63 h by gas chromatography.

Part 2. Effect of Water Addition on the Dialysis of a Mixture of Neutral, Acidic, and Basic Compounds. Three compositions were used: water-saturated diethyl ether, 1% and 2% (v/v) water/diethyl ether. The sample contained butanol, 2-octanone, tetradecane, octadecane, *n*-butyric acid, *n*-valeric acid, *n*-caproic acid, pyrazine, and 2,5-dimethylpyrazine, each at a concentration of 1 mg/mL. *n*-Nonyl acetate was incorporated as an internal standard. One milliliter of the diffusate was sampled after 72 h, dried over anhydrous sodium sulfate, and then analyzed via gas chromatography.

Gas Chromatographic Conditions. Except for the analyses of alcohols and fatty acids, a Hewlett-Packard Model 7620A research chromatograph equipped with a flame-ionization detector (FID) and a Hewlett-Packard 3390A integrator were used. Separation of compounds was accomplished on a 6 ft × 2 mm (i.d.) glass column packed with 10% Carbowax 20M on 100/120 Chromosorb W AW-DMCS (Supelco Co.). Nitrogen was used as the carrier gas at a flow rate of 30 mL/min. The injection port and detector temperatures were 250 and 280 °C, respectively. The column temperature was programmed from 100 to 170 °C at a rate of 8 °C/min with 1-min postinjection hold. A 2-μL sample was injected each time.

For the analysis of alcohols, a 6 ft × 2 mm (i.d.) glass column packed with 5% Carbowax 20M on 100/120 Chromosorb W AW-DMCS (Supelco Co.) was used. The oven temperature was programmed from 50 to 190 °C at a rate of 10 °C/min with a final hold for 1 min. The rest of the conditions were the same as described above.

For the analysis of fatty acids, a Hewlett-Packard 5880A gas chromatograph equipped with a flame-ionization detector was used. Separation of fatty acids was accomplished on a 30 m × 0.25 mm i.d. SE-54 fused silica bonded phase capillary column (J & W Scientific Co.). Hydrogen was used as the carrier gas with a column head pressure of 20 psi, which corresponded approximately to a linear velocity of 34 cm/s. The injection port and detector temperatures were 225 and 285 °C, respectively. The column temperature was first programmed from 40 to 100 °C at a rate of 30 °C/min with a 2-min postinjection hold and then programmed from 100 to 180 °C at a rate of 20 °C/min. Sample (0.3–0.4 μL) was injected in splitless mode.

RESULTS AND DISCUSSION

Continuing the research initiated by Benkler and Reineccius (1979, 1980), continuous dialysis was explored to isolate flavor constituents in this investigation. In the 40-m tubing type dialyzer designed, 1100 cm^2 of contact area was available with a surface to volume ratio of 46 prior to soaking the membrane in solvents. The dialysis rate was noticeably increased as compared to the previous batch method, and the dialysis time was reduced to 1–2 h from 24–72 h. This not only resulted in substantial saving of isolation time but also minimized the chances of artifact formation.

One of the problems initially confronted was swelling of the membrane in the solvents. This resulted in folding of the dialysis tubing and eventually obstructed the flow after limited usage of the system. Efforts were made to circumvent this problem, and it was found most effective to presoak the tubing in the solvents and then stretch the wet dialysis tubing in the dialysis unit. Extra tubing was then cut off before other ancillary parts were attached to the system.

Countercurrent flow of sample and solvent was chosen since higher concentration gradients could be accomplished and maintained for flavor dialysis. Flow rates of sample and solvent are the two major operating factors for a

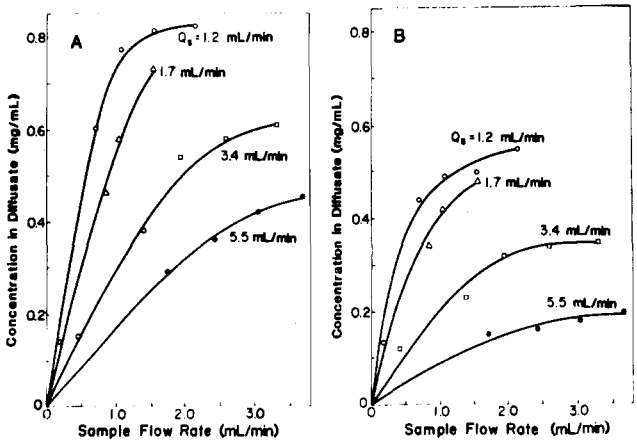

Figure 2. Effect of sample flow rate and solvent flow rate (Q_S) on the concentration in the diffusate: (A) butanol; (B) 2-octanone.

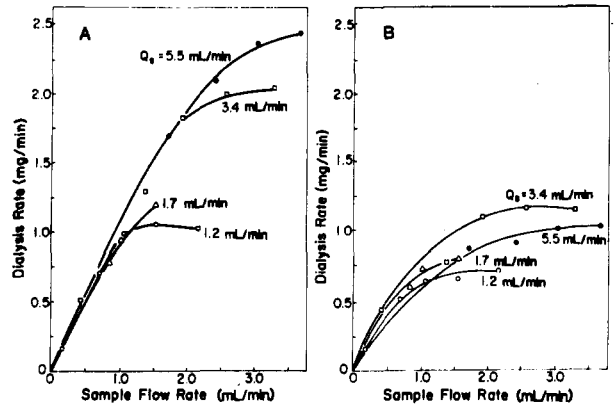

Figure 3. Effect of sample flow rate and solvent flow rate (Q_S) on the dialysis rate: (A) butanol; (B) 2-octanone.

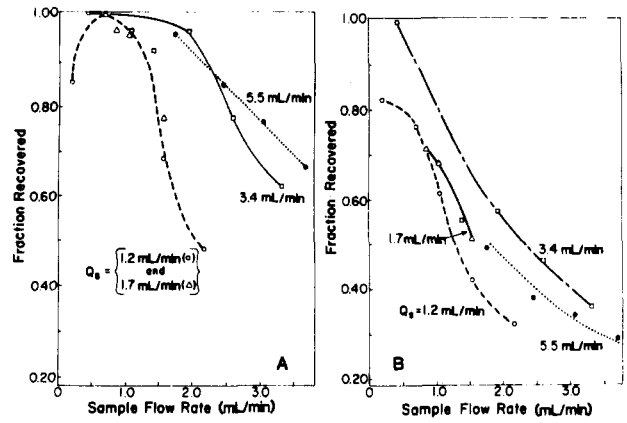

Figure 4. Effect of sample flow rate and solvent flow rate (Q_S) on the fraction recovered in the diffusate: (A) butanol; (B) 2-octanone.

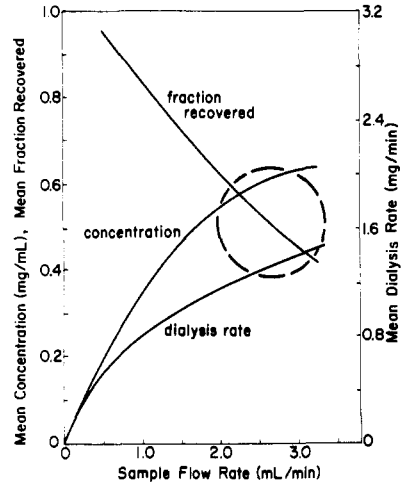

Figure 5. Interdependence of three parameters of dialysis efficiency at different sample flow rates for butanol and 2-octanone (solvent flow rate 1.7–3.4 mL/min).

continuous dialysis system. The effect of the flow rates on dialysis efficiency will be discussed for two representative compounds, butanol and 2-octanone.

A. Concentration. At constant solvent flow rates, diffusate concentrations increased with sample flow rates until a maximum was achieved (Figure 2). The slope of the linear part of the curve and maximum concentration achieved decreased when the solvent flow rates were increased. The observation that higher sample flow rates and lower solvent flow rates increased concentrations in the diffusates was expected. The higher sample flow rates would maintain higher average concentrations in the sample tube while the lower solvent flow rates permit longer contact time to accomplish equilibrium.

A comparison of diagrams A vs. B in Figure 2 shows that the maximum diffusate concentration observed for butanol was at 0.82 mg/mL and that for 2-octanone was only 0.55 mg/mL. The concentration of 2-octanone in the diffusate was generally less than that observed for butanol. This can be explained by the fact that molecular size is a primary determinant in the dialysis process; that is, the larger the molecule, the slower the diffusion rate.

B. Dialysis Rate. At constant solvent flow rates, the increase in sample flow rates facilitated dialysis rates of compounds until a maximum was reached (Figure 3). The positive effect of solvent flow rates on dialysis rates of butanol was observed only when the solvent flow rates were above 1.0 mL/min. A similar effect was found on 2-octanone. However, the optimal solvent flow rate for 2-octanone was at 3.4 mL/min instead of 5.5 mL/min, the highest rate employed. This can probably be explained by the fact that higher solvent flow rates not only provide a faster mass transport rate but also decrease contact time between sample and solvent. The effect of a faster mass transport rate may be diminished by lack of contact time when too high solvent flow rates were used for 2-octanone. Consequently, it resulted in the shift of optimal solvent flow rate. In light of the above discussion, it can be concluded that maximization of dialysis rates can be accomplished by maximizing both flow rates when the test compounds are relatively small molecules. For larger molecules, an optimal solvent flow rate needs to be established to maximize dialysis rates. The interaction between the flow rate effect and the molecular size effect of compounds makes the prediction of dialysis rates difficult.

C. Fraction Recovered. Figure 4 illustrates the influence of sample and solvent flow rates on recoveries of compounds. Fractions of compounds recovered increased with decreased sample flow rates and increased solvent flow rates. This is expected since longer contact time of sample with solvent and a faster mass transport rate in the solvent tube occurred under these conditions. However, increasing solvent flow rates above a certain point was unfavorable for the recovery of 2-octanone for reasons explained in part B. By maintaining the sample flow rates at a minimum (0.7–1.0 mL/min) and the solvent flow rates in the optimal region (3.4 mL/min), nearly complete recoveries of both compounds can be achieved.

The discussion so far indicates that changes in flow rates of sample and/or solvent had different impacts on concentration, dialysis rate, and recovery of flavor compounds in the diffusate. Figure 5 illustrates the interdependence

Table I. Diffusivities of Selected Neutral Compounds in the Flow Rate Effect Experiment

compd	slope, mL/min	r^2	diffusivity × 10^5, cm/s
butanol	6.06	0.84	9.18
2-octanone	1.67	0.84	2.54
tetradecane	ND[a]	<0.60	ND
2-decanone	0.87	0.72	1.32
nonanol	1.38	0.87	2.09
decanol	0.75	0.63	1.14
octadecane	ND	<0.60	ND

[a] ND = not determined due to poor linearity (r^2 < 0.60).

Table II. Effect of Methanol Concentration in the Solvent (Anhydrous Diethyl Ether) on Concentration of Flavor Compounds in the Diffusate (mg/mL) by Continuous Dialysis

compd	concn of methanol (v/v) 0%	1%	5%
hexanal	0.264	0.187	0.043
butanol	0.570	0.595	0.638
2-octanone	0.262	0.313	0.406
tetradecane	0.011	0.017	0.34
propionic acid	0.337	0.355	0.364
nonanol	0.207	0.252	0.311
benzyl acetate	0.185	0.235	0.324
octadecane	0	0	0

of these three measurements at different sample flow rates and essentially summarizes Figures 2–4. Such data could be used in providing a guideline for choosing the operating parameters of the dialysis system, depending on the ultimate purpose of the experiment. This would indicate that high recoveries cannot be achieved without sacrificing or compromising higher concentrations and faster rates. An instance where 100% recovery becomes essential would be quantitative analysis of flavors. When quantification is secondary, higher rates could be achieved to facilitate faster isolation (and may minimize chances of artifact formation) along with higher concentrations in the diffusate. For qualitative dialysis experiments, a compromise could be made among the three measures of dialysis efficiency. This would be the flow rates corresponding to the intersection of the three curves (circled region in Figure 5). Thus, a sample flow rate of 2–3 mL/min and a solvent flow rate of 1.7–3.4 mL/min are recommended in routine dialyses of flavor compounds with low molecular weight using this system.

A linear relationship between dialysis rates (R_D) and logarithmic mean concentration differences of sample and solvent (ΔC_{LM}) was found for all of the test compounds except tetradecane and octadecane (Table I). Tetradecane and octadecane could not be satisfactorily dialyzed and responded only slightly to the flow changes, due to the incompatible nature between these two compounds (hydrophobic) and the membrane (hydrophilic). This relationship indicated that the diffusivity of a given compound increased with increased polarity and decreased molecular

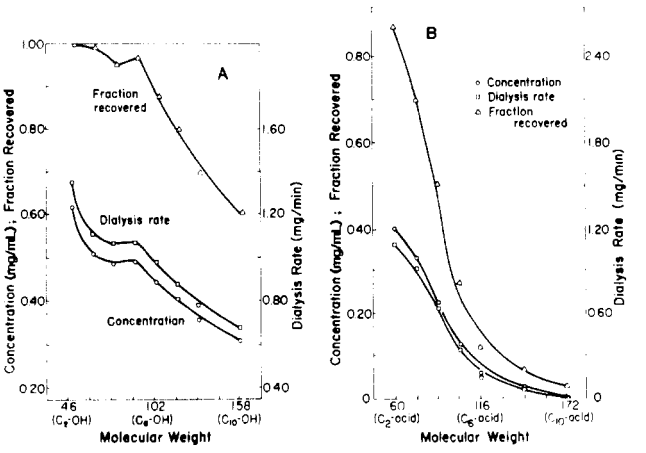

Figure 6. Effect of molecular weight on dialysis efficiency in a continuous dialysis system: (A) alcohols; (B) fatty acids.

size. The differences in diffusivity could be of 1 order of magnitude or more. Hypothetically, there are two opposing processes during diffusion of a compound, viz., partition process between solution and membrane and retention process within the membrane. These two opposing processes are governed by the chemical nature of the compound and the membrane. The net magnitude resulting from these two processes determines the overall observed diffusion rate. Since a polar membrane is used, the partition process probably predominates and leads to higher diffusivity for more polar compounds as observed.

The effect of molecular weight on dialysis efficiency is more clearly illustrated by Figure 6. An increase in molecular weights of both alcohols and fatty acids noticeably decreased dialysis efficiency except in the region of C_3–C_5 alcohols. In spite of the increase in molecular weight, the dialysis efficiency was not substantially different in this region. It is postulated that the polarity effect of the alcohols competed with the molecular weight effect. The retention effect due to polarity difference could be so predominant that the effect of molecular weight is not observable to any appreciable extent. The effect of molecular weight of fatty acids on dialysis efficiency diminished with an increase in molecular weight as observed from the change in slopes. Figure 6 also shows that, for an alcohol and fatty acid of the same carbon number, the alcohol could be dialyzed more easily than the fatty acid. This became more evident with higher molecular weight compounds. Since alcohols have smaller molecular dimensions and greater linearity in structure than the fatty acids, the observed result is expected.

The addition of methanol to anhydrous diethyl ether was found to improve diffusion of compounds across the membrane (Table II). The addition of methanol could have either dilated the pores of the membrane (Yeo et al., 1981) or increased the polarity of the membrane. Methanol concentration higher than 5% in anhydrous diethyl either (v/v) could not be employed for this system due to

Table III. Effect of Ammonium Hydroxide Content in the Solvent (Anhydrous Diethyl Ether) on the Batch Dialysis of Basic Compounds

compd	ammonium hydroxide content (v/v) 0% C_D[b]	0% C_{SS}	1% C_D	1% C_{SS}	2% C_D	2% C_{SS}	5% C_D	5% C_{SS}
pyrazine[a]	0	1.840	0.771	1.066	0.897	0.977	0.695	0.808
2-methylpyrazine	0	1.802	0.662	1.195	0.872	0.953	0.726	0.866
2,4,6-trimethylpyridine	0	1.649	0.239	1.682	0.687	1.121	0.581	1.123

[a] Sample was dialyzed for 72 h by batch process. [b] C_D = concentration in diffusate (mg/mL); C_{SS} = concentration in spent sample (mg/mL).

Table IV. Effect of Water Content in Solvent (Diethyl Ether) on the Batch Dialysis of a Mixture of Neutral, Acidic, and Basic Compounds[a]

compd	water content (v/v)		
	satd[b]	1%	2%
butanol	0.401	0.499	0.409
2-octanone	0.149	0.274	0.203
tetradecane	0.000	0.006	0.005
octadecane	0.000	0.000	0.000
propionic acid	0.278	0.344	0.245
n-butyric acid	0.310	0.278	0.285
n-valeric acid	0.264	0.299	0.277
n-caproic acid	0.095	0.141	0.103
pyrazine	0.226	0.435	0.336
2,5-dimethylpyrazine	0.277	0.295	0.222

[a] Diffusate was analyzed after 72 h. Concentration in mg/mL. [b] At 20 °C, water-saturated diethyl ether contains 1.2% water (w/w), which is approximately equivalent to 0.85% water (v/v).

excessive swelling of the dialysis tubing and subsequent obstruction of solvent flow. Although methanol was used in this investigation, it is not recommended for a real system considering the potential for artifact formation via aldol condensation reactions in presence of aldehydic components. Relatively low recoveries of hexanal (Table II) would indicate this possibility.

An effective method for enhancing the recovery of basic compounds was the addition of ammonium hydroxide to the solvent (Table III). The addition of 2% ammonium hydroxide (v/v) was found to be the most beneficial. However, approximately 10% of 2,4,6-trimethylpyridine was permanently lost. The addition of ammonium hydroxide can be employed only for acid-free samples since the potential for ammonium salt formation with acidic constituents exists and may impair dialysis of such compounds.

Since ammonium hydroxide contained a large proportion (ca. 70%) of water, water was suspected of being responsible for facilitating the dialysis of basic compounds rather than the ammonia itself. Table IV shows that neutral, acidic, and basic compounds could all be dialyzed in the presence of water. The optimal water content in the solvent was 1% (v/v). The concentration of test compounds in the diffusate decreased beyond 1% (v/v) water content. This may be due to an unfavorably large increase in the membrane polarity with excess water. A plausible explanation for the membrane performance is that ammonium hydroxide and/or water may serve as a swelling agent and dilate the membrane pores. However, further investigation is needed to elucidate the exact mechanism. Water, therefore, may be added directly to the samples containing neutral, acidic, and basic compounds to facilitate the dialysis process while ammonium hydroxide may be used only for acid-free samples. The above-discussed results obtained by employing the batch dialyzer were directly translated to a continuous system as well, in other experiments not presented herein.

ACKNOWLEDGMENT

We thank Professor E. L. Cussler, Department of Chemical Engineering and Materials Science, University of Minnesota, Minneapolis, for useful discussions and S. Knoblauch for typing this manuscript. This paper was presented at the 43rd Annual Meeting of the Institute of Food Technologists, New Orleans, LA, June 19–22, 1983. Published as Paper No. 14,488 of the scientific journal series of the Minnesota Agricultural Experiment Station on research conducted under Minnesota Agricultural Experiment Station Project No. 18-83.

Registry No. Butanol, 71-36-3; 2-octanone, 111-13-7; tetradecane, 629-59-4; 2-decanone, 693-54-9; nonanol, 28473-21-4; decanol, 112-30-1; octadecane, 593-45-3; ethanol, 64-17-5; propanol, 71-23-8; pentanol, 71-41-0; hexanol, 111-27-3; heptanol, 111-70-6; octanol, 111-87-5; acetic acid, 64-19-7; propionic acid, 79-09-4; butyric acid, 107-92-6; valeric acid, 109-52-4; caproic acid, 142-62-1; caprylic acid, 124-07-2; capric acid, 334-48-5; hexanal, 66-25-1; benzyl acetate, 140-11-4; pyrazine, 290-37-9; 2-methylpyrazine, 109-08-0; 2,4,6-trimethylpyridine, 108-75-8; 2,5-dimethylpyrazine, 123-32-0; diethyl ether, 60-29-7; ammonium hydroxide, 1336-21-6.

LITERATURE CITED

Arnold, R. G.; Barnhart, H. M. *J. Dairy Sci.* **1972**, *55*, 1069.
Benkler, K. F.; Reineccius, G. A. *J. Food Sci.* **1979**, *44*, 1525.
Benkler, K. F.; Reineccius, G. A. *J. Food Sci.* **1980**, *45*, 1084.
Buttery, R. G. *J. Agric. Food Chem.* **1973**, *21*, 31.
Chang, S. S.; Vallese, F. M.; Hwang, L. S.; Hsieh, O. A. L.; Min, D. B. S. *J. Agric. Food Chem.* **1977**, *25*, 450.
Grot, W. *Chem. Ing. Technol.* **1972**, *4*, 167.
Honkanen, E.; Karvonen, P. *Acta Chem. Scand.* **1966**, *20*, 2626.
Jennings, W. G.; Rapp, A. "Sample Preparation for Gas Chromatographic Analysis"; Hüthig Verlag: Heidelberg, New York, 1983; Chapter 3, p 33.
Michaels, A. S. *Trans.*—Am. *Soc. Artif. Intern. Organs.* **1966**, *12*, 387.
Reineccius, G. A. In "Modern Methods of Food Analysis"; Stewart, K. K., Whitaker, J. R., Eds.; AVI Publishing Co.: Westport, CT, 1984; Chapter 12, p 293.
Reineccius, G. A.; Anandaraman, S. In "Food Constituents and Food Residues: Their Chromatographic Determination"; Lawrence, J. F., Ed.; Marcel Dekker: New York, 1984; Chapter 5, p 195.
Schreier, P. "Chromatographic Studies of Biogenesis of Plant Volatiles"; Hüthig Verlag: Heidelberg, New York, 1984; Chapter 1, p 1.
Yeo, R. S.; Chan, S. F.; Lee, J. *J. Membr. Sci.* **1981**, *9*, 273.

Received for review March 29, 1985. Accepted July 18, 1985.