Analysis of Wine Aroma by Direct Injection in Gas Chromatography without Previous Extraction

Jesús Villén,† Francisco J. Señoráns,‡ Guillermo Reglero,‡ and Marta Herraiz*,‡

Facultad de Ciencias Químicas, Universidad de Castilla-La Mancha, Campus Universitario s/n, 13004 Ciudad Real, Spain, and Instituto de Fermentaciones Industriales, CSIC, c/Juan de la Cierva 3, 28006 Madrid, Spain

A method is proposed which allows the direct gas chromatographic analysis of real-life samples of wine (i.e., without the need of preliminary sample preparation) just by using a programmed temperature vaporizer injector operated in a modified solvent elimination mode. The procedure presented is ecologically beneficial as no solvent is required for its performance; it is not expensive since it does not demand a special methodology; and it might be used as a quick screening in several fields. A comparison of the proposed method with other isolation techniques (i.e., continuous liquid–liquid extraction and simultaneous distillation–extraction) usually employed for the concentration of volatile components is also included.

Keywords: *Wine aroma; isolation and concentration techniques; direct injection; programmed temperature vaporizer (PTV)*

INTRODUCTION

In past years the necessity for new sample preparation techniques has been often underscored and different procedures suitable for the chromatographic analysis of trace compounds have been reported (Jennings and Rapp, 1983; Poole and Schuette, 1983). Isolation and concentration techniques are mainly used to obtain more concentrated samples, but the possibility of taking out interfering substances and simultaneously improving the detectability of specific solutes is very interesting for the posterior chromatographic analysis.

It is generally admitted, however, that no single sample preparation procedure is best suited for all samples under all conditions. That is why the knowledge of the advantages and disadvantages of each specific method is strongly recommended, in order to select the most adequate to enhance the analytical capabilities of the experimentation to be performed.

Liquid–liquid extraction has been previously used for obtaining an efficient enrichment during the isolation of volatile compounds (Hardy, 1969; Rapp et al., 1976; Brander et al., 1980; Fagan et al., 1982; Etievant and Bayonove, 1983), but it is a time-consuming procedure which requires the concentration of the solutes prior to the chromatographic analysis itself. This step can occasionally not only cause the loss of some volatile compounds, but also produce eventual interferences resulting from solvent impurities which may significantly decrease the obtained recoveries. Recently, Ferreira et al. (1993) have developed a fast method for analyzing wine volatiles based on a microextraction with Freon 113. Evidently, the use of minimum solvent/sample volume ratios enhances concentration and minimizes contamination problems, but differences in solvent–solvent partition coefficients may seriously affect the relative amounts recovered.

Trichlorofluoromethane (Freon 11) has previously been recommended as a suitable solvent for liquid–liquid extraction of food flavors, particularly those having high alcoholic contents such as alcoholic beverages (Hardy, 1969; Rapp et al., 1976).

The simultaneous distillation–solvent extraction technique (SDE) (Likens and Nickerson, 1964; Nickerson and Likens, 1966; Godefroot et al., 1981, 1982) has also been found to be advantageous for an effective concentration of different types of food samples using small amounts of extractive solvents, without requiring a further concentration step (Núñez and Bemelmans, 1984; Núñez et al., 1984; de Frutos et al., 1988; Blanch et al., 1991, 1992). However, the temperature required to perform the process may eventually be a serious problem for accomplishing the reliable analysis of thermolabile compounds.

On the other hand, the use of a programmed temperature vaporizer (PTV) has proved to be versatile for the reliable injection of complex mixtures in gas chromatography, avoiding discrimination sources related to hot sampling (Schomburg, 1981; Poy et al., 1981). This system has also been found suitable for achieving an adequate enrichment of volatile components just by packing the glass liner of the injector body with a suitable material (Poy et al., 1982; Nitz and Jülich, 1984; Nitz et al., 1984; Poy and Cobelli, 1985; Herraiz et al., 1987, 1989; Tabera et al., 1991). Also the concentration of an extract previously obtained by liquid–liquid extraction inside the PTV (internal concentration) has been already successfully carried out (Villén et al.,1989; Loyola et al., 1990).

In previous works we have proposed the use of experimental designs to optimize the introduction of large sample volumes in capillary gas chromatography by using a PTV (Villén et al., 1992; Señoráns et al., 1993). Recently, we have also evaluated the use of the mentioned injector operated in a modified solvent split mode for performing the direct injection of a wine distillate made from Muscat grapes (Pisco). This sampling procedure offers evident advantages for the analysis of real-life samples, mainly concerning the short time required to perform the overall analysis and its reliability.

Nevertheless, up to now the isolation and concentration of wine aroma compounds have been carried out

† Universidad de Castilla-La Mancha.
‡ Instituto de Fermentaciones Industriales.

0021-8561/95/1443-0717$09.00/0 © 1995 American Chemical Society

almost exclusively by using continuous liquid–liquid extraction. The possibility of introducing the sample directly into the gas chromatograph without performing any kind of pretreatment is of evident interest for the analysis of minor volatile compounds. However, such a procedure has not been applied so far to wine analysis because the eventual thermal decomposition inside the injector of nonvolatile compounds might produce the occasional elution of ghost peaks which would render the experimentation difficult.

The aim of this work was to investigate the feasability of using a PTV injector in a modified solvent split mode (solvent elimination) for performing the direct analysis of wine without previous sample preparation. A comparison of the proposed method with those involving the continuous liquid–liquid extraction and the simultaneous distillation and extraction of the sample is also included.

EXPERIMENTAL PROCEDURES

Sample. A wine from the Verdejo cultivar (Rueda, Valladolid, Spain) having an alcoholic content equal to 12% (v/v) was used for all experiments. The aroma of Verdejo wine is considered to be fresh, fruity, and fragrant and very different from the aroma resulting from other Spanish grape varieties (Herraiz et al., 1991). In fact, the choice of the mentioned cultivar was mainly due to its complex volatile composition which seemed to be adequate to comparatively evaluate different isolation and concentration methods.

Capillary Gas Chromatography-Mass Spectrometric Analysis. All samples were analyzed by gas chromatography using a 50 m × 0.22 mm i.d. fused-silica capillary column (SGE, Ringwood, Australia) coated with a 0.25 μm layer of cross-linked BP-21.

Analyses were carried out in a Perkin-Elmer gas chromatograph (Model 8310) equipped with a PTV injector and a FID operated at 250 °C. The equipment was coupled to a Model 2600 chromatography software system (Nelson Systems, Inc.). Helium was used as carrier gas (36 psig). In all instances, the temperature of the gas chromatographic column was first kept at 35 °C for 8 min and then programmed at 2 °C/min to 180 °C.

To confirm peak identification, the gas chromatograph was linked to a Perkin-Elmer ITD-50 ion trap detector (electron impact, 70 eV). The comparison of spectra data with those of the NBS (National Bureau of Standards) library provided us with initial information of component identities, and supplementary verification was achieved in some cases by matching their spectral data with those of reference compounds analyzed under identical conditions.

Liquid–Liquid Extraction. Two hundred fifty milliliters of wine was continuously extracted for 24 h in downward displacement extractors with 150 mL of double-distilled Freon 11 which had been purchased from Atochem. The extractors were kept cold by using a cryogenic unit.

As internal standard, a 0.2-μL volume of methyl octanoate was added to the extract which was subsequently either concentrated before being sampled into the gas chromatograph (external GC–concentration) or injected into the gas chromatograph without previous concentration (internal GC–concentration). Experimentation was performed as described below.

Liquid–Liquid Extraction with External Concentration (LLE-EC). The Freon extract obtained was concentrated by means of a 50-cm length of Vigreux column maintained at 32 °C in a water bath and subsequently analyzed by gas chromatography using a PTV operated in the cold split mode (sampled volume, 2 μL; split ratio, 50:1). The body injector was maintained at 40 °C while sampling, then increased (at 14 °C/s) to 375 °C and maintained for 5 min. In all instances the injections were carried out by discharging the syringe (a 10-μL SGE syringe, Model R-GP) quickly into the injector.

Liquid–Liquid Extraction with Internal GC Concentration (LLE-IC). The internal GC concentration of the extracts previously obtained by liquid–liquid extraction was carried out by trapping in a suitable adsorbent placed in the glass liner of the PTV injector which was operated in the solvent elimination mode.

In this work a 2-cm plug of Tenax TA (80/100 mesh) was packed in the vaporization insert (90 mm × 1 mm i.d. × 2 mm o.d.). For injections in the solvent split mode, the PTV was held at 30 °C upon injection (split ratio, 50:1). The split valve was closed at the end of the trapping step (30 s) and the PTV was ballistically heated to 350 °C for 5 min. In order to avoid a possible injection overflow, the syringe (a 25-μL SGE syringe, Model R-GP) was discharged slowly. In all cases the sampled volume was 25 μL. Values established for different experimental variables such as the type of adsorbent, split ratio, initial and end PTV temperatures, and the time of trapping were selected according to our previous experience (Herraiz et al., 1987; Villén et al., 1989).

Simultaneous Distillation–Extraction. Isolation and concentration of the compounds of interest was performed by means of the micro version of the distillation–extraction device proposed by Godefroot et al. (1981) in the configuration which allows the use of extraction solvents denser than the sample solvent.

The SDE extraction of the aroma compounds of the wine investigated was satisfactorily achieved after having diluted the sample until an alcoholic content equals 3% (what involves a dilution of 1:4 v/v).

In all cases a 250-mL of the diluted wine sample was heated by applying an oil bath at 120 °C, whereas a 2-mL volume of the solvent (dichloromethane) distills by heating with a water bath at 50 °C. The sample and solvent volumes were established to fill the return arms of the SDE apparatus and to achieve the required demixing equilibrium. A cold finger refrigerated at −5 °C allowed the vapors of solvent and sample to be condensed. The preconcentration step was considered to be complete in 90 min.

Methyl octanoate was also used as internal standard by adding a 0.2-μL volume to the SDE extract obtained. Subsequently the gas chromatographic analysis was performed in two different operation modes as described below.

Simultaneous Distillation–Extraction with Concentration (SDEC). The GC analysis of the SDE extract was performed after having concentrated the sample under a stream of inert gas.

A 10-μL syringe SGE Model R-GP was used and the injection was accomplished in the cold splitless mode (sampled volume, 2 μL).

Simultaneous Distillation–Extraction without Concentration (SDENC). In this case, a 2-μL volume of the SDE extract was sampled into the chromatograph without carrying out a previous concentration. The PTV was now operated in the cold splitless mode and a syringe SGE, Model R-GP, was used.

Direct Injection (DI). The direct analysis of wine aroma did not require the previous extraction and concentration of the compounds of interest. The method which we propose for the analysis of real-life samples only demands the use of a PTV injector operated in a modified solvent split mode.

The injection was accomplished by using a simple, inexpensive, homemade device which had been previously proposed to allow the introduction of large sample volumes by using a syringe (SGE, Model A-RN, 500 μL) in such a way that the speed of sample introduction can be carefully controlled.

The vaporization insert of the PTV injector was packed with a 4.5-cm length plug of an adsorbent containing 55% GasChrom 220 and 45% Tenax TA which had been previously washed with acetone and subsequently conditioned under a stream of nitrogen either 30 min at 90 °C, 30 min at 180 °C, 60 min at 300 °C, and 120 min at 350 °C (for Tenax TA) or 30 min at 90 °C, 30 min at 180 °C, and 120 min at 250 °C (for GasChrom 220). The packed liners were conditioned for 120 min at 255 °C under a purge flow rate of helium.

Experimental conditions for the direct analysis of wine aroma were selected according to our previous experience

Table 1. Comparative Estimation of the Extraction Efficiency Obtained by Direct Injection (DI), Liquid–Liquid Extraction with External Concentration (LLE-EC), Liquid–Liquid Extraction with Internal Concentration (LLE-IC), Simultaneous Distillation–Extraction with Concentration (SDEC), and Simultaneous Distillation–Extraction without Concentration (SDENC)

		DI	LLE-EC		LLE-IC		SDEC		SDENC	
no.	compound	*a*	*a*	*b*	*a*	*b*	*a*	*b*	*a*	*b*
1	ethyl lactate + *cis*-3-hexen-1-ol	29.21	24.16	82.70	21.07	72.14	8.04	27.54	7.12	24.38
2	acetic acid	19.60	0.92	4.68	0.39	1.98	0	0	0	0
3	propanoic acid	1.64	0.62	37.50	0.75	45.89	0.33	20.36	0.23	13.90
4	linalool	64.50	1.34	2.08	1.07	1.67	0	0	0	0
5	γ-butyrolactone	2.91	1.18	40.56	1.07	36.75	0	0	0	0
6	butanoic acid	0.24	0.29	118.01	0.23	93.17	0	0	0	0
7	α-terpineol	13.54	14.65	108.24	14.27	105.42	1.53	11.32	1.32	9.78
8	hexanoic acid	1.80	2.66	147.88	2.19	121.72	0.08	4.26	0	0
9	benzyl alcohol	1.04	1.77	170.03	1.61	154.33	0	0	0	0
10	2-phenylethanol	27.91	69.93	250.57	66.23	237.33	0.87	3.12	1.01	3.62
11	octanoic acid	0.55	0.29	53.05	0.25	45.75	0	0	0	0
12	ethyl hexadecanoate	0.28	0.61	220.83	0.75	270.48	0	0	0	0
13	decanoic acid	43.24	0.55	1.27	0.13	0.30	0	0	0	0

[a] Extraction efficiency expressed as average of the ratio of the absolute peak area corresponding to each peak to that for standard ($n = 6$). [b] Extraction efficiency given as percentage of that one obtained by direct injection.

Table 2. Relative Standard Deviations ($n = 6$) for the Absolute and Relative (Normalized) Peak Areas for the Analysis of Wine during Direct Injection (DI), Liquid–Liquid Extraction with External Concentration (LLE-EC), Liquid–Liquid Extraction with Internal Concentration (LLE-IC), Simultaneous Distillation–Extraction with Concentration (SDEC), and Simultaneous Distillation–Extraction without Concentration (SDENC)

		DI		LLE-EC		LLE-IC		SDEC		SDENC	
no.	compound	absolute areas	normalized areas	absolute areas	normalized areas	absolute areas	normalized areas	absolute areas	normalized areas	absolute areas	normalized areas
1	ethyl lactate + *cis*-3-hexen-1-ol	4.5	14.6	38.5	8.7	15.8	8.5	16.1	9.2	14.4	10.8
2	acetic acid	16.3	13.6	40.2	8.6	15.9	12.0				
3	propanoic acid	8.1	12.9	34.5	4.4	20.8	18.1	12.7	5.7	17.0	11.9
4	linalool	10.1	8.7	37.6	6.5	17.2	11.9				
5	γ-butyrolactone	14.7	3.0	39.2	11.8	19.8	14.7				
6	butanoic acid	16.2	13.2	41.9	8.5	20.6	9.1				
7	α-terpineol	10.7	17.4	38.0	5.1	10.8	3.7	15.5	11.2	14.7	8.3
8	hexanoic acid	15.5	20.7	31.7	13.7	13.3	5.0	28.7	32.8		
9	benzyl alcohol	7.4	14.3	30.5	2.7	14.4	3.9				
10	2-phenylethanol	5.7	10.2	38.0	5.5	11.7	3.4	20.2	17.6	14.8	16.9
11	octanoic acid	25.1	11.8	30.4	12.7	13.8	3.1				
12	ethyl hexadecanoate	12.6	7.0	20.1	24.2	11.3	5.1				
13	decanoic acid	19.8	6.8	49.2	20.5	12.9	4.9				

concerning the introduction of large sample volumes in a gas chromatograph by means of a PTV (Señoráns et al., 1993). Before sampling, the capillary column was disconnected from the injector body while simultaneously maintaining a flow through the vaporizing chamber of 597 mL/min. In this way, condensation of the solvent inside the column was prevented. The initial temperature of the PTV was kept at 9 °C over the injection period. The system was maintained in the mentioned conditions for 5 min after the sampling; the carrier gas flow was then switched off and the column was again connected to the injector. Subsequently the carrier gas was turned on and the PTV temperature was ballistically raised (14 °C/s) to 250 °C so that the desorption step allowed the transference of the solute material to the capillary column (splitless mode). The end temperature of the PTV injector was kept for 5 min. After having finished the analysis, the capillary column was again disconnected, the split valve was reopened and the PTV was heated at 250 °C for 5 min to allow the effective purge of the vaporizing chamber.

In all instances the sampled volume was 50 μL with 0.18 μL/s being the speed of sample introduction which was established to be the proper injection speed with respect to the solvent evaporation rate.

Methyl octanoate was used as external standard for quantitative calculations. To this aim a 2-μL volume of a solution in dichloromethane (20 μL/L) was injected in the cold splitless mode.

For each series of measurements, the reported values were calculated from a minimum of six replicate injections.

RESULTS AND DISCUSSION

Data obtained by the different methods which have been evaluated are collected in Tables 1 and 2. Table 1 gives a comparative estimation of the extraction efficiencies of the investigated techniques by considering the ratio of the absolute peak area corresponding to each compound to that for standard. As can be seen, values resulting from the direct injection of the sample, liquid–liquid extraction with external concentration, liquid–liquid extraction with internal concentration, simultaneous distillation–extraction with concentration, and simultaneous distillation–extraction without concentration are included. The values (as percentages), resulting from the ratio of the average extraction efficiency obtained by using each one of the investigated techniques to that one achievable for direct injection for each compound, are also given.

It is clear that the direct injection technique provides extraction efficiencies much higher than the simultaneous distillation–extraction does, while a further comparison with data corresponding to the liquid–liquid extraction (both with internal and external GC concentration) does not reveal such differences.

The relative standard deviations (RSD) resulting from both the absolute and relative (normalized) peak areas are given in Table 2. The lowest values are, in general, obtained if the wine is analyzed by direct injection

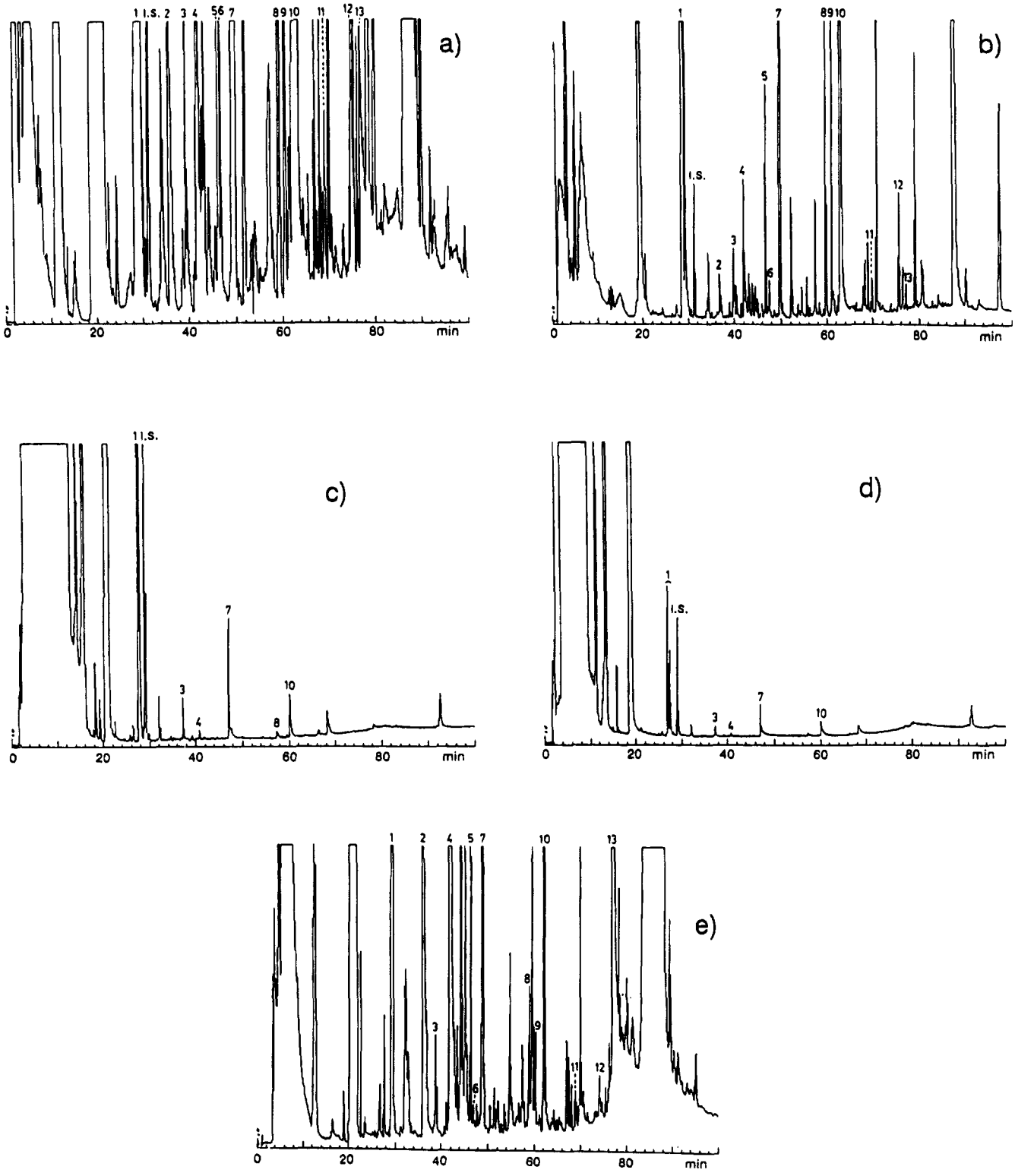

Figure 1. Chromatograms from the aroma of a wine of the Verdejo variety obtained by (a) liquid–liquid extraction with external concentration, (b) liquid–liquid extraction with internal concentration, (c) simultaneous distillation–extraction with concentration, (d) simultaneous distillation–extraction without concentration, and (e) direct injection. (See text for further details.) (Identification peak numbers are as in Table 1.)

specially when absolute peak areas are used for calculations. As far as the liquid–liquid extraction is concerned, it is evident that the internal concentration of the extract should be preferred to the external concentration.

Figure 1 shows the chromatograms corresponding to the analysis of the Verdejo wine investigated by the five different methods considered. It is interesting to note that all the mentioned chromatograms were recorded at the same full range so that it is possible to state that the SDE procedure clearly produces a loss of volatiles compounds (Figure 1d) which is less evident if a concentration of the SDE extract is performed prior to its sampling into the gas chromatograph (Figure 1c).

It is worthy to note that the sample volume (50 μL) allowed the detection of the investigated compounds, but the possibility of dramatically improving the sensibility achievable in the direct analysis of wine aroma just by

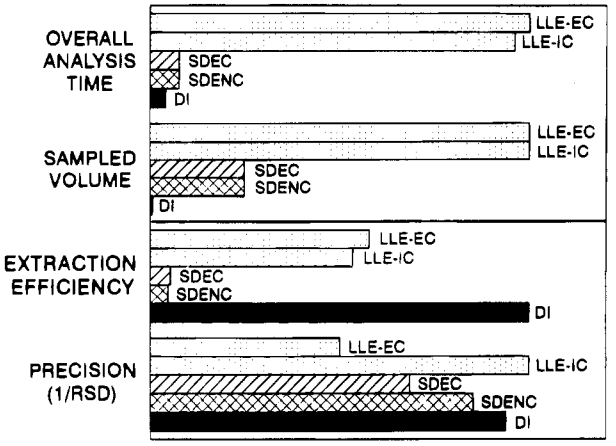

Figure 2. Comparison of different isolation and concentration techniques for the GC analysis of wine aroma in terms of analysis time, sampled volume, extraction efficiency, and precision.

increasing the sample volume should be emphasized. In our laboratory, up to at least 485 μL have been previously introduced into a capillary column without producing column overflooding (Señoráns et al., 1993) so that this approach could be of interest for analyzing trace compounds of sensorial relevance or responsible of an eventual adulteration or anomaly of wine aroma.

Figure 2 gives a comparison of the investigated methods of sample concentration in terms of the analysis time required for the overall procedure, the sampled volume, the estimated extraction efficiency, and the precision achievable in each case. The direct injection procedure proposed in this work for the analysis of wine aroma seems to be the most adequate as it provides a complete information, in a very short time (approximately 2 h), from a very small amount of sample (50 μL) without requiring any kind of sample pretreatment. Moreover, the extraction efficiency and precision of the overall analysis (expressed in Figure 2 as the average values resulting, respectively, from extraction efficiencies and the reciprocal of relative standard deviations corresponding to the investigated compounds) are quite satisfactory in comparison to these one resulting from the liquid–liquid extraction and the simultaneous distillation–extraction.

A further aspect to be considered is the eventual decomposition of thermally unstable compounds during the desorption step. In this respect, it is interesting to emphasize that the spectrometric analysis did not evidence the occurrence of artifacts in the chromatograms obtained by direct injection of the investigated wine since all the identified compounds are typically found in the aroma of alcoholic beverages.

LITERATURE CITED

Blanch, G. P.; Reglero, G.; Herraiz, M.; Tabera, J. A comparison of different extraction methods for the volatile components of grape juice. *J. Chromatogr. Sci.* **1991**, *29*, 11–15.

Blanch, G. P.; Tabera, J.; Sanz, J.; Herraiz, M.; Reglero, G. Volatile composition of vinegars. Simultaneous distillation-extraction and gas chromatographic-mass spectrometric analysis. *J. Agric. Food Chem.* **1992**, *40*, 1046–49.

Brander, C. F.; Kepner, R. E.; Webb, A. D. Identification of some volatile compounds of wine of vitis vinifera, cultivar Pinor noir. *Am. J. Enol. Vitic.* **1980**, *31*, 69–75.

de Frutos, M.; Sanz, J.; Martínez Castro, I. Simultaneous distillation-extraction (SDE) method in the qualitative and quantitative GC analysis of cheese volatile components. *Chromatographia* **1988**, *25*, 861–64.

Etievant, P. X.; Bayonove, C. L. Aroma components of pomaces and wine from the variety Muscat de Frontignan. *J. Sci. Food Agric.* **1983**, *34*, 393–403.

Fagan, G. L.; Kepner, R. E.; Webb, A. D. Additional volatile components of Palomino film sherry. *Am. J. Enol. Vitic.* **1982**, *33*, 47–50.

Ferreira, V.; Rapp, A.; Cacho, F.; Hastrich, H.; Yavas, I. Fast and quantitative determination of wine flavor compounds using microextraction with freon 113. *J. Agric. Food Chem.* **1993**, *41*, 1413–20.

Godefroot, M.; Sandra, P.; Verzele, M. New method for quantitative essential oil analysis. *J. Chromatogr.* **1981**, *203*, 325–35.

Godefroot, M.; Stechele, M.; Sandra, P.; Verzele, M. A new method for the quantitative analysis of organochlorine pesticides and polychlorinated biphenyls. *HRC & CC* **1982**, *5*, 75–79.

Hardy, P. J. Extraction and concentration of volatiles from dilute aqueous and aqueous-alcoholic solution using trichlorofluoromethane. *J. Agric. Food Chem.* **1969**, *17*, 656–58.

Herraiz, M.; Reglero, G.; Loyola, E.; Herraiz, T. Sampling of volatile components using a PTV in the solvent split mode. *HRC & CC* **1987**, *10*, 598–602.

Herraiz, M.; Reglero, G.; Herraiz, T. Evaluation of a PTV injector for quantitative analysis of volatile compounds at low concentrations. *HRC* **1989**, *12*, 442–46.

Herraiz, T.; Reglero, G.; Martín-Alvarez, P. J.; Herraiz, M.; Cabezudo, M. D. Identification of aroma components of Spanish "Verdejo" wine. *J. Sci. Food Agric.* **1991**, *55*, 103–16.

Jennings, W. G.; Rapp, A. *Sample preparation for gas chromatographic analysis*; Bertsch, W., Jennings, W. G., Kaiser, R. E., Eds.; Hüthig: Heidelberg, 1983.

Likens, S. T.; Nickerson, G. B. Detection of certain hop oil constituents in brewing products. *Am. Soc. Brew. Chem. Proc.* **1964**, 5–13.

Loyola, E.; Martín-Alvarez, P. J.; Herraiz, T.; Reglero, G.; Herraiz, M. A contribution to the study of the volatile fraction in distillates of wines made from muscat grapes (Pisco). *Z. Lebensm. Unters. Forsch.* **1990**, *190*, 501–05.

Nickerson, G. B.; Likens, S. T. Gas chromatographic evidence for the occurrence of hop oil components in beer. *J. Chromatogr.* **1966**, *21*, 1–5.

Nitz, S.; Jülich, E. Concentration and GC-MS analysis of trace volatiles by sorption-desorption techniques. In *Analysis of volatiles*; Schreier, P., Ed.; de Gruyter: New York, 1984; pp 151–70.

Nitz, S.; Drawert, F.; Jülich, E. Trace analysis by intermediate trapping with a modified programmable temperature vaporizer. *Chromatographia* **1984**, *18*, 313–17.

Núñez, A. J.; Bemelmans, J. M. H. Recoveries from an aqueous model system using a semi-micro steam distillation-solvent extraction procedure. *J. Chromatogr.* **1984**, *294*, 361–65.

Núñez, A. J.; Bemelmans, J. M. H.; Maarse, H. Isolation methods for the volatile components of grapefruit juice. Distillation and solvent extraction methods. *Chromatographia* **1984**, *18*, 153–58.

Poole, C. F.; Schuette, S. A. Isolation and concentration techniques for capillary column gas chromatographic analysis. *HRC & CC* **1983**, *6*, 526–49.

Poy, F.; Cobelli, L. Programmed temperature vaporizer (PTV) injection. In *Sample introduction in capillary gas chromatography*; Sandra, P., Ed.; Hüthig: Heidelberg, 1985; Vol. 1, pp 77–97.

Poy, F.; Visani, S.; Terrosi, F. Automatic injection in high-resolution gas chromatography: A programmed temperature vaporizer as a general purpose injection system. *J. Chromatogr.* **1981**, *217*, 81–90.

Poy, F.; Visani, S.; Terrosi, F. A universal sample injection system for capillary column GC using a programmed temperature vaporizer (PTV). *HRC & CC* **1982**, *5*, 355–59.

Rapp, A.; Hastrich, H.; Engel, L. Gas cromatographic investigations on the aroma constituents of grape. I. Concentration and separation by capillary glass columns. *Vitis* **1976**, *15*, 29–36.

Schomburg, G. Sampling systems in capillary chromatography. *Proceed. 4th Int. Symp. on Capillary Chromatography. Hindelang*; Kaiser, R. E., Ed.; Hüthig: Heidelberg, 1981; pp 371–74.

Señoráns, F. J.; Tabera, J.; Villén, J.; Herraiz, M.; Reglero, G. Variables affecting the introduction of large sample volumes in capillary gas chromatography using a programmed-temperature vaporizer. *J. Chromatogr.* **1993**, *648*, 407–14.

Tabera, J.; Reglero, G.; Herraiz, M.; Blanch, G. P. Optimization of dynamic headspace sampling for the analysis of trace volatile components of grape juice: Use of a PTV injector for intermediate trapping. *HRC* **1991**, *14*, 392–96.

Villén, J.; Herraiz, T.; Reglero, G.; Herraiz, M. Experiments with the PTV in the solvent split mode for concentration of volatiles. *HRC* **1989**, *12*, 633–35.

Villén, J.; Señoráns, F. J.; Herraiz, M.; Reglero, G.; Tabera, J. Experimental design optimization of large volume sampling in a programmed temperature vaporizer. Application in food analysis. *J. Chromatogr. Sci.* **1992**, *30*, 261–66.

Received for review August 8, 1994. Revised manuscript received November 18, 1994. Accepted December 28, 1994.[⊗] This work was made possible by financial assistance from the DGICYT, Project PB91-0077-C03-03. F.J.S. thanks the Ministerio de Educación y Ciencia for a grant.

JF940452T

[⊗] Abstract published in *Advance ACS Abstracts,* February 15, 1995.