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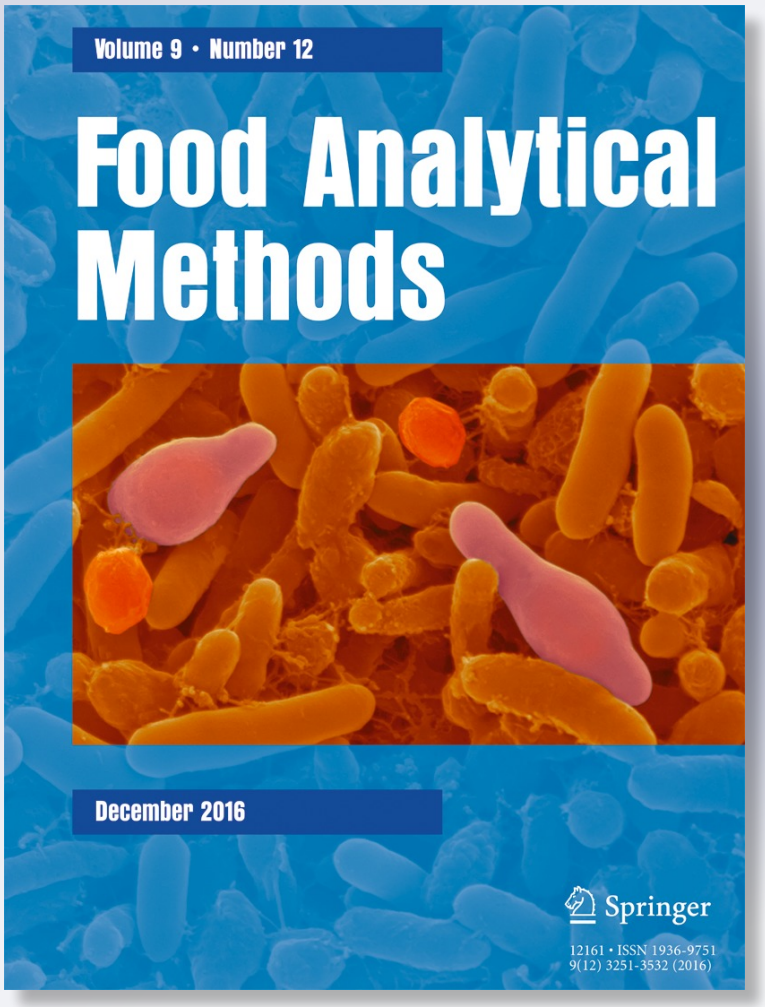

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QuEChERS Method for the Determination of 3-Alkyl-2-Methoxypyrazines in Wines by Gas Chromatography-Mass Spectrometry

Ariel R. Fontana[1] · **Rubén Bottini**[1]

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Abstract A new method for determining 3-isopropyl-2-methoxypyrazine, 3-*sec*-butyl-2-methoxypyrazine, and 3-isobutyl-2-methoxypyrazine in wines is presented. A modified quick, easy, cheap, effective, rugged, and safe method and gas chromatography-mass spectrometry (GC-MS) were used for sample preparation and compound determination, respectively. The analytes were extracted from wine (30 mL) with 1 mL toluene, in the presence of 12 g anhydrous $MgSO_4$ and 3 g NaCl. Cleanup of the toluene phase was performed by a miniaturized dispersive solid-phase extraction with a combination of anhydrous $CaCl_2$ (25 mg), anhydrous $MgSO_4$ (25 mg), and primary-secondary amine (10 mg), which was effective for minimizing co-extractives and matrix effects. GC-MS parameters were also tuned up to optimize limits of detection between 4.2 and 7.1 ng L^{-1}. The overall recoveries (trueness) of the method ranged between 71 and 87 % for the white and red wine samples, respectively, spiked at 40 and 100 ng L^{-1}, with relative standard deviations below 21 %. The method was applied for the determination of target methoxypyrazines in the samples of commercial wines from Argentina.

Keywords Methoxypyrazines · Wine · QuEChERS · Dispersive solid-phase extraction · Sample preparation · Gas chromatography-mass spectrometry

✉ Ariel R. Fontana
afontana@mendoza-conicet.gob.ar; fontana_ariel@yahoo.com.ar

[1] Laboratorio de Bioquímica Vegetal, Instituto de Biología Agrícola de Mendoza, Consejo Nacional de Investigaciones Científicas y Técnicas-Universidad Nacional de Cuyo, Almirante Brown 500, M5528AHB Chacras de Coria, Argentina

Introduction

Alkyl methoxypyrazines (MPs) are nitrogen-containing heterocyclic molecules that are aroma active compounds found in many wine varietals. These compounds may be grape-derived or introduced into wines via grape-associated microorganisms (Allen et al. 1991; Buchbauer et al. 2000; Kögel et al. 2014). MPs in wine contribute to green, herbaceous, and vegetal aroma notes associated with specific odors of bell peppers, green peas, beets, potatoes, and asparagus (Allen et al. 1991; Botezatu et al. 2014; Roujou de Boubee et al. 2000; Sidhu et al. 2015). Notwithstanding their origin, MPs may be beneficial for wine quality, contributing to the specificity of certain wine varietals (Cabernet Sauvignon, Cabernet Franc, Sauvignon Blanc). At higher concentration levels, MPs have a detrimental role resulting in overpowering green, unripe, and herbaceous notes giving a negative connotation to wines (Dunlevy et al. 2013; Hein et al. 2009). The three MPs 3-isobutyl-2-methoxypyrazine (IBMP), 3-isopropyl-2-methoxypyrazine (IPMP), and 3-sec-butyl-2-methoxypyrazine (SBMP) are the most conspicuous and studied in wines. The IBMP is the predominant pyrazine present in wine representing approximately 80 % of the 5 to 50 ng L^{-1} found in some wines (Allen and Lacey 1997; Hartmann 2003). In this sense, due to the relevant organoleptic impact of MPs on wine quality and authenticity at very low concentration levels, the development of analytical methodologies for the determination of them in wine samples is a subject of growing interest.

Due to the complexity of wine matrix (high quantity of analytes with different chemical nature and concentration levels) and the low expected concentration of target MPs (ng L^{-1} range), sample preparation plays an important role in their reliable determination. In this way, diverse sample preparation approaches have been proposed being headspace solid-phase microextraction (HS-SPME) and solid-phase

extraction (SPE) the most used (Botezatu et al. 2014; Godelmann et al. 2008; Kotseridis et al. 2008; Legrum et al. 2014; López et al. 2011; Prouteau et al. 2004; Sala et al. 2002; Sidhu et al. 2015). Both techniques reported good recoveries and suitable limits of detection (LODs) for such levels of MPs found in wines, being adequate alternatives from this point of view. However, these methods have various limitations including lengthy and multistep procedure, high analysis cost, poor selectivity, and high degree of matrix effect.

Recently, simplification and miniaturization of extraction systems with the aim to increase sample throughput and reduce solvent consumption (and consequently wastes) have gained interest in the analytical chemistry field. In 2003, Anastassiades et al. (2003) reported the QuEChERS (quick, easy, cheap, effective, rugged, and safe) sample preparation technique for the analysis of multi-residual pesticides in fruits and vegetables. A salting-out step for extraction of analytes coupled with the novel dispersive solid-phase extraction (d-SPE) cleanup avoids multiple extraction steps as well as passing the extracts through SPE cartridges, requiring much smaller quantities of sorbent and solvent. By simplifying and/or integrating some classical sample preparation methods, QuEChERS reduces the amount of sample used and provides significant saving of reagents, materials, energy, and time required for the analysis (Dmitrienko et al. 2014). Due to these advantages, it was applied to analytes and matrices of different chemical nature, showing excellent results in terms of analytical performance and simplification of quantification in complex matrices. Up to now, applications of QuEChERS in wine matrixes have been developed for determination of pesticides (Romero-González et al. 2011), mycotoxins (Pizzutti et al. 2014), phthalic acid esters (Xu et al. 2014), and phenolic compounds (Fontana and Bottini 2014) coupled to liquid chromatography with different detectors. Patil et al. (2011) developed a QuEChERS method to extract 2,4,6-trichloroanisole from wines prior to GC-MS, showing distinct advantages in terms of economy and time of analysis as well as reproducible results even after analyzing a lot of samples. However, to the best of our knowledge, the QuEChERS method has not been optimized and applied for the extraction of MPs from wines.

For the determination step, gas chromatography-mass spectrometry (GC-MS)-based techniques have been the choice for MP determination, providing efficient preconcentration techniques are used to achieve sensitivity and selectivity (Sidhu et al. 2015). Recently, new strategies based on multidimensional GC-MS and GC × GC have been proposed for quantifying MPs in juices and grapes with the aim to avoid problems related to sample complexity (Botezatu et al. 2014).

The objective of this work was to develop a simple, fast, efficient, precise, and cheap sample preparation approach for the determination of MPs in white and red wines by GC-MS. Determination by GC-MS with single ion recording (SIR) mode and QuEChERS sample preparation conditions were optimized in order to maximize the sensitivity and selectivity of the methodology. The performance of the proposed method was evaluated in terms of LODs, limits of quantification (LOQs), recoveries, and linear working range. Applicability of the methodology was evaluated by analyzing different commercial wines from Argentina.

Materials and Methods

Standards, Solvents, and Sorbents

Standards of MPs were purchased from Sigma-Aldrich (Steinheim, Germany) and consisted of the following: 2-isobutyl-3-methoxypyrazine (IBMP, ≥99 % purity), 2-isopropyl-3-methoxypyrazine (IPMP, 99 % purity), and 2-sec-butyl-3-methoxypyrazine (SBMP, 97 % purity). Stock solutions of the above MPs were prepared in absolute ethanol at a concentration of 2000 μg mL^{-1}. Intermediate dilutions were prepared in ethanol (for wine spiking) or toluene (calibration standards) depending on the final concentration required and stored in the dark at −20 °C. The calibration standards were prepared by successive dilutions.

Toluene, ethyl acetate, and hexane were from Merck (Darmstadt, Germany) of HPLC grade. Formic acid (FA) was purchased from Mallinckrodt Baker (Inc. Phillipsburg, NJ, USA). Analytical grade sorbents (50-μm particle size) for d-SPE, including primary-secondary amine (PSA) and octadecylsilane (C_{18}), were both obtained from Waters (Milford, MA, USA). Reagent grade NaCl, sodium hydroxide (NaOH), $MgSO_4$, and $CaCl_2$ for QuEChERS development were purchased from Biopack (Buenos Aires, Argentina).

Samples

Extraction conditions were optimized with aliquots of a pool of red wines from the Malbec variety, spiked with target analytes at 1 ng mL^{-1}. The same blend was used for matrix effect and recovery experiments. For this study in white wines, a blend without varietal denomination was used.

Wine samples analyzed in this work were obtained from local supermarkets and from a winery. The studied samples included different white and red wines produced in Argentina. The white wines corresponded to Sauvignon Blanc variety and red wines were from Cabernet Sauvignon and Merlot varieties. Each wine sample was analyzed in triplicate, and concentration results were expressed as the average concentrations (ng L^{-1}) with their standard deviations.

Sample Preparation

Wine (30 mL) was placed into a 50-mL PTFE centrifuge tube with 0.6 mL NaOH (500 g L^{-1} solution), 1 mL toluene was

added, and the tube was vigorously hand-shaken for 1 min to ensure adequate homogenization. For phase separation, 12 g of $MgSO_4$ and 3 g of NaCl were added; the tubes were hand-shaken for 1 min and centrifuged for 10 min at 8000 rpm. The upper organic layer along with a portion of the matrix (2 mL) was collected in a 10-mL centrifuge tube and further centrifuged for 3 min. The supernatant was collected in a 0.8-mL Eppendorf tube and cooled 15 min at −20 °C. The extract was immediately cleaned up by d-SPE with a mixture of anhydrous $CaCl_2$ (25 mg), anhydrous $MgSO_4$ (25 mg), and PSA (10 mg), then vortexed for 1 min, followed by centrifugation at 8000 rpm for 6 min. Finally, a 2-μL aliquot of the cleaned extract was injected in splitless mode into the GC-MS for identification and quantification of MPs.

Chromatographic Conditions

GC-MS analyses were carried out on a Clarus 500 capillary gas chromatograph coupled to a single-quadrupole mass spectrometer detector (Perkin Elmer, Shelton, CT, USA). The GC column used was an HP-5MS (30 m × 0.25 mm, 0.25-μm film thickness) (Agilent Technologies, Wilmington, DE, USA). The temperature program was 60 °C, ramped at 5 °C min^{-1} to 110 °C (1-min hold), then at 20 °C min^{-1} up to 230 °C, and finally at 40 °C min^{-1} to 300 °C with a hold for 3 min. The injector temperature was set at 240 °C, and the injections were carried out in the splitless mode. The mass spectrometer was operated in electron impact ionization mode at 70 eV. The transfer line and ion source temperatures were maintained at 200 and 180 °C, respectively. The samples were analyzed in SIR mode. The peak identification was based on the base peak and the isotopic pattern of the MPs. Specific ions were selected for each MP, and the base ion was selected as a quantitative ion. Quantifier ions were 137, 138, and 124 m/z for IPMP, SBMP, and IBMP, respectively, while qualifier ions were 152 for IPMP, 138 for SBMP, and 151 for IBMP. Two SIR segments were used, one for IPMP (8–9 min) with a dwell time of 0.21 s (137 and 152 m/z) and the other segment for SBMP and IBMP (9.5–11 min) with a dwell time of 0.18 s (138, 124, and 151 m/z). The LODs of the target compounds were set at a signal-to-noise (S/N) ratio of 3, whereas the LOQs were set at S/N of 10 based on the five-point external calibration graph obtained using matrix-matched standards.

Matrix Effects, Absolute Recoveries, and Enhancement Factor

Potential matrix effects (MEs) for each compound caused by interferences occurring during GC-MS analysis were evaluated by comparing the slope of the calibration lines based on the matrix-matched standards of red and white wines with the slope of the solvent-based calibration lines. A higher slope of the matrix calibration indicates matrix-induced signal enhancements, whereas a lower slope represents signal suppressions.

The recovery experiment was carried out in red and white wines at concentration levels of 40 and 100 ng L^{-1} ($n = 3$). The samples were extracted according to the method described in the "Sample Preparation" section. Quantification was performed by external calibration using matrix-matched standards.

Enhancement factor (EF) was calculated as the ratio between the initial wine sample volume and the resulting toluene extract after applying the QuEChERS technique considering the obtained recoveries for MPs of each wine matrix.

Results and Discussion

Optimization of GC-MS Parameters

The ion source temperature was optimized to achieve the highest S/N for MPs. It was performed by stepwise temperature increments from 150 to 200 °C. Increases in S/N were observed by increasing the temperature up to 180 °C (i.e., 2800 to 4100 S/N values for SBMP). Higher temperatures do not report significant differences; thus, 180 °C was selected as optimum.

Dwell time was optimized to attain highest sensitivity in GC-MS with SIR mode. Initially, full scan MS of MPs were acquired to identify the target compounds and to choose the ions to be monitored according to their highest intensity. From the full scan spectra presented in Fig. 1, base peaks for IPMP, SBMP, and IBMP were 137, 138, and 124 m/z, respectively. These ions were selected for quantification whereas the ions 152 m/z for IPMP, 124 m/z for SBMP, and 151 m/z for IBMP were used as qualifiers. Two SIR scan functions were created, and dwell times were optimized for each, evaluating the peak area and S/N for MPs. The optimum dwell times were 0.21 s for the first segment with two ions (IPMP 137 and 152 m/z) and 0.18 s for the second, including three ions (SBMP and IBMP 124, 138, and 151 m/z). These values are approximately one tenth of the GC peak width at the base (using toluene as solvent), so giving about 10 points across the peak, which is the optimal condition to increase SIR sensitivity and GC peak area precision. Diminishing the dwell time causes a considerable decrease in S/N and peak area of MPs, with a reduction between 50 and 70 % of S/N. By augmenting the parameter over the selected values, the sensitivity decreases. So, the optimized dwell times give the best sensitivity and peak area precision for MPs under studied conditions.

Optimization of QuEChERS Extraction Conditions

The efficiency of MP extraction was affected by several factors, including the extraction solvent, its volume, and other

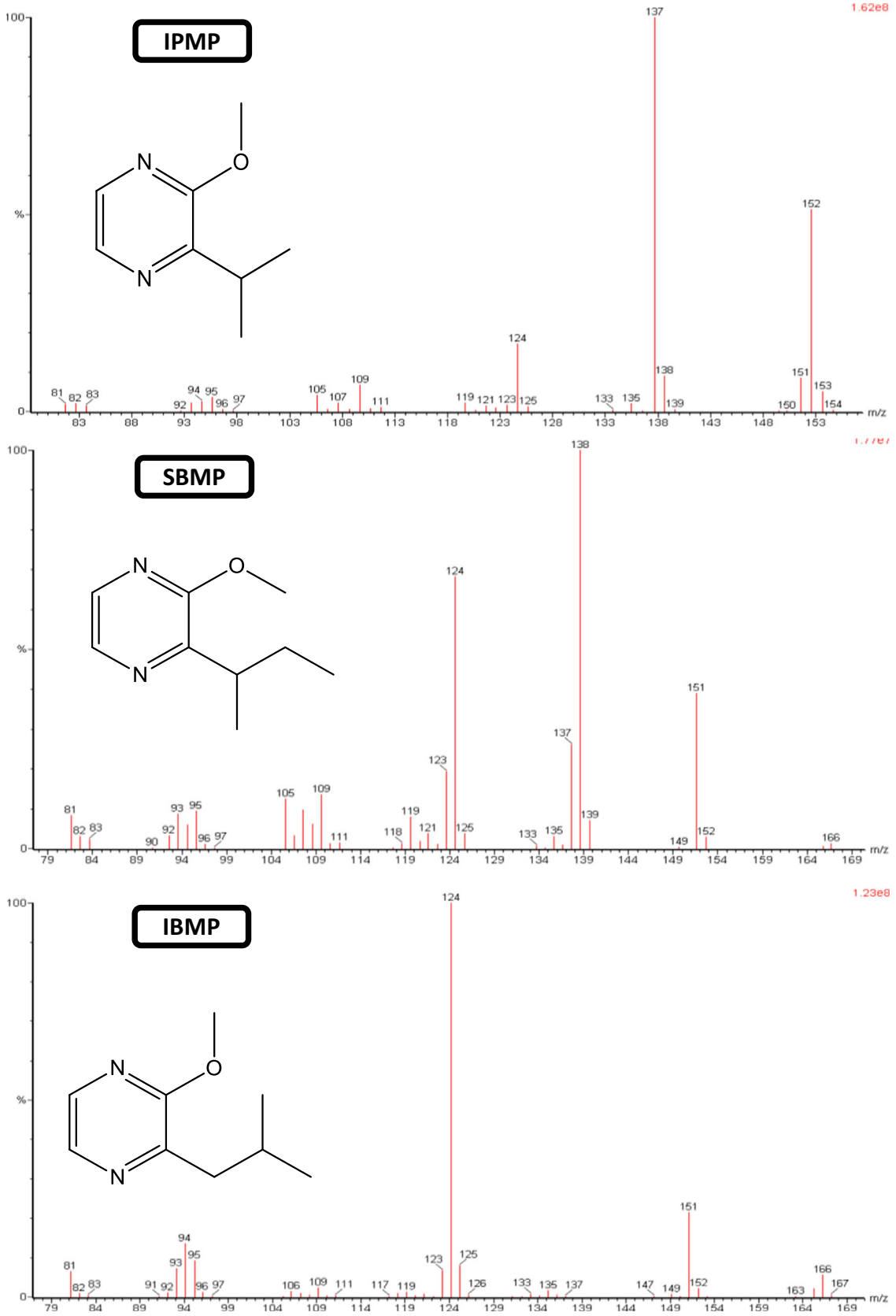

Fig. 1 Chemical structures and GC-MS full scan mass spectra of IPMP, SBMP, and IBMP

variables that influence the sample preparation technique such as pH. The study and optimization of these variables were performed by modifying one at a time while keeping the remaining factors unchanged.

The optimum extraction solvent was selected based on the solubility of MPs, selectivity (lower co-extraction of matrix components), and GC compatibility. Different extraction solvents were evaluated including toluene, hexane, and ethyl acetate. MPs showed the highest recoveries (between 21 and 27 %) by using toluene as solvent (Fig. 2a). As well, toluene presented higher S/N than hexane and ethyl acetate and also sharpened peaks. This could be related to the presence of matrix components extracted with these solvents, which was evidenced by a visual appearance of the corresponding extracts. For the same matrix, the most complex extracts corresponded to ethyl acetate, which rendered intense dark extracts. Toluene allowed obtaining colorless, completely transparent extracts. The volumetric recovery (0.75–0.85 mL out of 1 mL) was also highest for toluene followed by ethyl acetate and hexane (0.55–0.60 mL). To evaluate the transfer efficiency of MPs from the injector to the detector, peak width of analytes by using different solvents was evaluated (Fig. 2b). Toluene achieved narrowed peaks (between 30 and 50 %) and presented more uniform peak shapes with greater injection repeatability in comparison to the other solvents. An additional explanation to this result could be the higher boiling point of toluene (110 °C) in comparison to hexane (68 °C) and ethyl acetate (77 °C). This fact could allow a better solvent effect focusing which occurs for higher volatility analytes (i.e., MPs). Thus, toluene was selected as the extraction solvent for further studies.

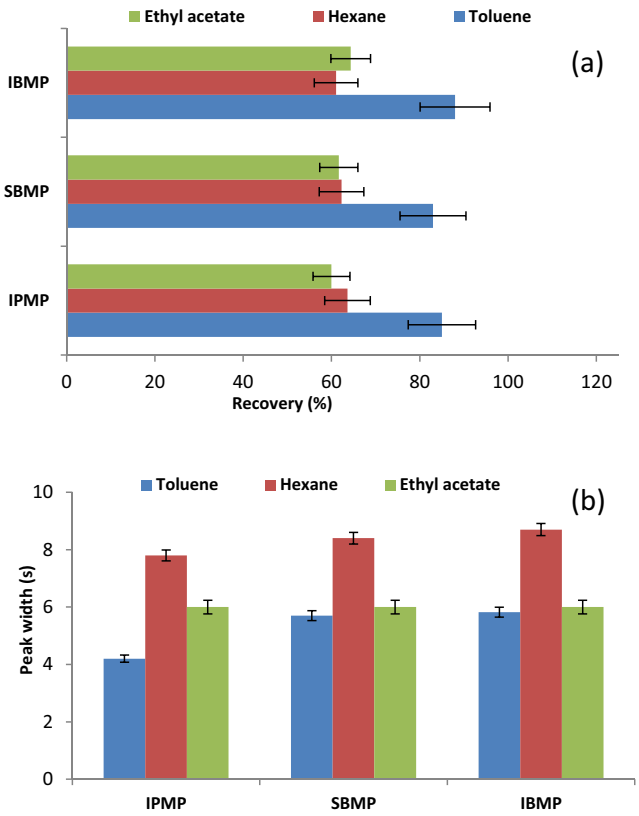

Fig. 2 **a** Effect of type of solvent on recovery of MPs. **b** Effect of solvent on the peak width of analytes. Extraction conditions: sample volume, 30 mL red wine; extraction solvent volume, 1 mL; addition level, 1 ng mL^{-1}. d-SPE 500 μL extract, 25 mg anhydrous $CaCl_2$, 25 mg $MgSO_4$, 10 mg PSA, 10 mg C_{18}

The sample pH adjustment prior to extraction is a commonly used strategy to increase the recovery of acidic or basic compounds. The MPs are organic bases, which are protonated at low pH forming quaternary ammonium ions. This characteristic makes the pH a critical parameter, because the addition of a basic solution to increase sample pH will increase the recovery of MPs from wines. At neutral or basic pH, the amine-ammonium equilibrium shifts towards the less polar pyrazine form and MPs are mostly neutral molecules, facilitating the extraction with non-polar organic solvents such as toluene. The pH effect on the MP extraction was evaluated by adding different concentrations of NaOH (0.5, 0.8, and 1.2 %, *w*/*v*) or FA (1 %, *v*/*v*) to the samples and compared with sample without pH modification (raw sample). Table 1 shows the results of the three MPs at different pH conditions. A significant increase in the responses of IPMP, SBMP, and IBMP can be seen as pH increases from 2 (1 % FA) to 6 (0.8 % NaOH). Further pH increase to 8 did not report significant differences in the responses for analytes. Additionally, the S/N achieved at pH 6 was much higher than the one obtained at lower pH, probably because at low pH more matrix co-extractives of wine are present as neutral analytes (i.e., phenolic compounds) remaining in toluene phase after the salting-out step. It was evidenced by a dark appearance of extracts. At pH 6 or higher, the extracts were colorless because most of the co-extractives from wine are present as ionic compounds, reducing their mass transfer to the non-polar toluene phase. Thus, the samples were adjusted at a pH value of approximately 6 by adding 0.8 % NaOH.

The sample-to-solvent ratio was studied with the objective to achieve the highest recoveries with the minimum sample

Table 1 Mean values of MP analytical responses (peak areas) at different pH conditions. Values ± standard deviations (triplicate measurements)

	IPMP	SBMP	IBMP
1 % FA	1424 ± 106	1767 ± 110	2115 ± 130
Raw wine	1286 ± 98	1905 ± 108	2456 ± 168
0.5 % NaOH	2366 ± 125	3671 ± 179	3996 ± 199
0.8 % NaOH	2962 ± 156	4584 ± 200	4567 ± 215
1 % NaOH	2860 ± 176	4550 ± 199	4515 ± 208

Extraction conditions: sample volume, 30 mL red wine; extraction solvent volume, 1 mL toluene; addition level, 1 ng mL^{-1}. d-SPE conditions as described in Fig. 2

and solvent consumption, as well as to get the required sensitivity for MPs in wines. To determine the influence of extraction solvent volume, a series of separate sets of extractions were performed using 30:1 and 40:1 ratios. The volumetric recovery of the toluene phase was in the range of 0.8–0.85 mL and 0.6–0.65 mL for the 30:1 and 40:1 ratios, respectively. Nearly 85 % of the solvent phase could be recovered for the sample volume of 30 mL. For sample volumes of 40 mL or higher, the volumetric recovery of the toluene phase was lower, with inefficient phase separation due to an emulsion formation at the interface which significantly affects the reproducibility of this step. As well, the obtained recoveries were lower probably because the reduced volumetric recovery of extraction phase was insufficient to quantitatively extract the target analytes. Taking this fact into consideration, a sample-to-solvent ratio of 30:1 was selected.

Optimization of d-SPE Cleanup

For the development of d-SPE, PSA and C_{18} were evaluated alone and in different combinations. In the same way, the addition of anhydrous $CaCl_2$ was studied taking into account that it has been reported as reducing the solubility of esters and fatty acids present in wines (decanoic acid, tetradecanoic acid, palmitic acid, etc., and their alkyl esters) on QuEChERS extracts (Patil et al. 2011). Removal of fatty acids is important to reduce the matrix effect and background noise, allowing the improvement of LODs. Due to the reduced volume of extract achieved during extraction step, a miniaturized cleanup was proposed by reducing the amount of sorbents used in d-SPE. The effectiveness of each sorbent was evaluated in terms of recoveries of the target analytes and background level of the chromatograms (assessed as S/N). These results were compared against those achieved with the QuEChERS extract without cleanup. The d-SPE procedure was as follows: 300 μL aliquot of extract was cleaned up with 10 mg of the selected d-SPE sorbent (or combinations) plus 25 mg $CaCl_2$ and 25 mg $MgSO_4$. After sorbent addition, the tube was vortexed and centrifuged. Afterwards, 75 μL aliquot of the cleaned extract was put in an autosampler vial and further analyzed by GC-MS. This approach allowed to use a low amount of extract (such thus obtained in our salting-out step) as well as to reduce the quantity of sorbents during cleanup, minimizing the input cost of sample processing for a single sample. The achieved results are summarized in Fig. 3. In terms of MP recoveries (Fig. 3a), the best results were obtained by using the combination of PSA, $CaCl_2$, and $MgSO_4$; also, extracts without cleanup or using $CaCl_2 + MgSO_4$ (without PSA and C_{18}) showed comparable recoveries with the former d-SPE approach. The use of C_{18} and the combination of C_{18} with PSA reported recovery values between 21 and 55 %, which are lower than the former condition. It may be because that at the working pH, the MPs are present as neutral analytes

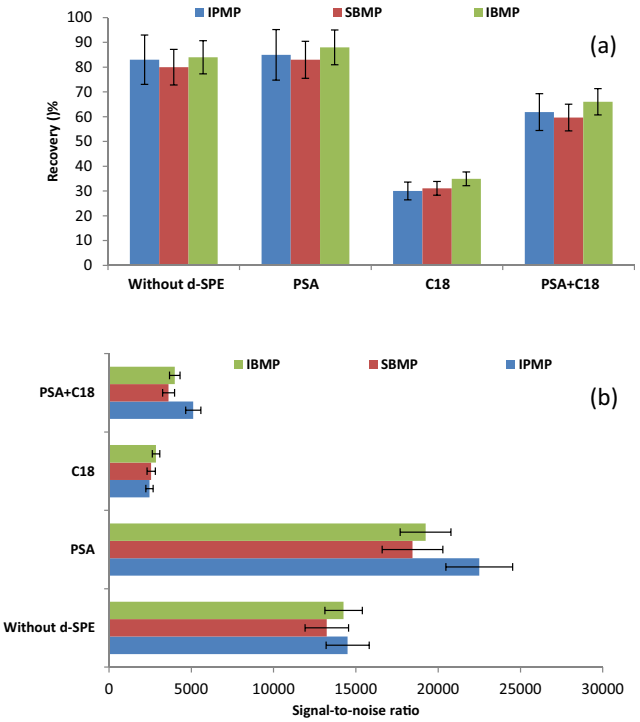

Fig. 3 **a** Effect of type of sorbent on the recoveries of MPs. **b** Effect of d-SPE sorbent on the S/N ratio of MPs. Extraction conditions as described in Fig. 2

being strongly retained in the non-polar C_{18}. At this pH, analytes cannot interact with PSA, so anhydrous $MgSO_4$ retains the remaining water after the salting-out step and increases the ionic strength of the medium, therefore improving the partition of neutral analytes to toluene phase. Additionally, the presence of $CaCl_2$ increases the ability of PSA to retain fatty acids, contributing to achieve cleaner chromatograms and mass spectra. Evaluating the results in terms of S/N of analytes (Fig. 3b), the results obtained showed that the use of a combination of the PSA, $CaCl_2$, and $MgSO_4$ gives the best results. The utilization of C_{18} alone or in combination with PSA showed the lowest S/N for MPs (77–89 % lower than the best condition). By omitting the cleanup step, the S/N were 25–35 % lower than by using the optimum d-SPE conditions, demonstrating the relevance of applying the cleanup strategy over crude extracts. In this way, the application of the proposed cleanup significantly lowered the peak area of the remaining fatty acid (143 m/z) and other co-extractives which allowed reducing the instrumental limits of quantification. Similar results were achieved by other authors in the analysis of wine samples (Patil et al. 2011).

Analytical Performance and Application to Wine Samples

In order to evaluate the ME on the analytical signals of MPs, the slopes of the calibration lines obtained with matrix-matched standards were compared with those obtained with

solvent-based standards, calculating the matrix to solvent slope ratios for each of the analytes studied in white and red wine matrices. Matrix-induced signal suppression was observed for IPMP in both wines, being up to 32 % in red and 30 % in white wines. Signal enhancements were observed for SBMP (47 and 50 % for red and white wines, respectively) and IBMP (28 and 14 % in red and white wines, respectively). Hence, considering different extents of ME, it is recommended to prepare separate matrix-matched standards for white and red wines. Since 30 mL wine volume was extracted in 0.80 mL toluene (volume finally recovered after QuEChERS extraction step) and taking into account the obtained average recoveries of the method for both white and red wines (Table 2), the EFs obtained were nearly 30. The calibration curves were constructed with five levels of concentration in triplicate. For both calibration curves, linear ranges between 20 and 100 ng L^{-1} were obtained for the studied MPs (see Table 2). The LODs of the analytes for extraction of 30 mL wine sample, calculated as three times the signal-to-noise ratio (S/N = 3), were 6.4, 7.1, and 4.2 ng L^{-1} for IPMP, SBMP, and IBMP, respectively. The LOQs, calculated as 10 times the signal-to-noise ratio (S/N = 10), were 19.3, 23.6, and 13.9 ng L^{-1} for IPMP, SBMP, and IBMP, respectively.

The recovery of MPs, considered as an estimation of trueness, was evaluated at two concentration levels (40 and 100 ng L^{-1}). In all cases, spiked and non-spiked aliquots were processed in triplicate and the concentrations of MPs in the corresponding extracts determined by matrix-matched calibration. The achieved recoveries were within the range of 71–87 % with the RSDs ($n = 3$) between 17 and 21 % for the low level and 9 and 14 % for the high level. The obtained results are summarized in Table 2. The achieved RSD values may be satisfactory considering the low concentration levels at which these compounds were evaluated, being good evaluations of the method trueness.

The developed and validated QuEChERS-GC-MS method was applied for the determination of MPs to a total of five samples of white (three specimens) and red (two specimens) wines from different grape varieties cultivated in Argentina. As was mentioned previously, several differences were observed between the white and red wines and; thus, quantification should be performed by a matrix-matched calibration curve to ensure accurate results. Different samples of commercial wines were analyzed, but the studied MPs were not detected. Although it may be possible that MPs were below the detection limit of the proposed methodology, most probably the analytes had been not present. In fact, MP levels in wines depend on several factors previously reported such as light exposure, crop level, and vintage between others (Dunlevy et al. 2013; Legrum et al. 2014; Ryona et al. 2008). Legrum et al. (2014) informed about not detectable levels of MPs for a given vintage; accrediting this results in a general climatic influence, meaning that MPs are considerably lower in some years than in others.

Conclusions

A simple, robust, and low-cost sample preparation method has been proposed as a convenient alternative for determining trace levels of MPs in wine samples by GC-MS. Under optimized working conditions, the developed methodology provides suitable recoveries and linear response ranges. The proposed QuEChERS-GC-MS offers distinct advantages over the conventional sample preparation techniques. It shows a lower organic solvent consumption than most SPE methodologies (1 versus 7–10 mL), and it is considerably faster than the time-consuming HS-SPME. Additional sake compared to SPME is the possibility of processing several samples simultaneously, improving the sample throughput and usefulness of the method in screening studies involving the analysis of many samples in a short time. In this way, the proposed method had definite virtues in terms of lower input cost and time of analysis. In addition, the application of the cleanup strategy achieves cleaner extracts, being expected an increase in GC liner and column life-time, summed to the miniaturization of d-SPE that allows reducing the amount of required sorbents. Altogether, the above features guarantee the ruggedness of the proposed methodology for the routine screening of target MPs in commercial wine samples, with the aim of evaluating their organoleptic influence on quality and authenticity of wines.

Table 2 Analytical parameters and recoveries of the QuEChERS-GC-MS method for the determination of MPs in white and red wines

	Linear range (ng L^{-1})	LOD (ng L^{-1})	LOQ (ng L^{-1})	Recovery (%) ± RSD, $n = 3$ replicates, 40 ng L^{-1}		Recovery (%) ± RSD, $n = 3$ replicates, 100 ng L^{-1}	
				White wine	Red wine	White wine	Red wine
IPMP	20–100	6.4	19.3	78 ± 21	81 ± 20	79 ± 13	84 ± 14
SBMP	25–100	7.1	23.6	75 ± 19	71 ± 17	85 ± 12	82 ± 11
IBMP	20–100	4.2	13.9	79 ± 18	80 ± 19	83 ± 10	87 ± 9

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Compliance with Ethical Standards

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Conflict of Interest Ariel R. Fontana declares that he has no conflict of interest. Rubén R. Bottini declares that he has no conflict of interest.

Ethical Approval This article does not contain any studies with human participants or animals performed by any of the authors.

Informed Consent Not applicable.

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