Multi-residue method to determine selected personal care products from five classes in fish based on miniaturized matrix solid-phase dispersion and solid-phase microextraction coupled to gas chromatography-mass spectrometry

Iran Ocaña-Rios,[a] Bhawana Thapa,[a] and Jared L. Anderson[a*]

[a]*Department of Chemistry, Iowa State University, Ames, Iowa 50011, USA.*

Abstract

A method featuring matrix solid-phase dispersion combined with solid-phase microextraction coupled to gas chromatography-mass spectrometry was developed to determine parabens, musks, antimicrobials, UV filters, and an insect repellent in fish. Optimization and validation of the method was carried out on tilapia and salmon samples. Acceptable linearity ($R^2 > 0.97$), precision (relative standard deviations < 13 %) and accuracy (recovery > 80 %) at two concentration levels for all analytes were obtained using both matrices. The limits of detection ranged from 0.01 to 1.01 μg g^{-1} (wet weight) for all analytes except for methyl paraben. The SPME Arrow format was applied to increase the sensitivity of the method, and yielded detection limits more than ten times lower than those achieved with traditional SPME. The miniaturized method can be applied to various fish species, regardless of their lipid content, and represents a useful tool for quality control and food safety purposes.

Corresponding Author:
Jared L. Anderson
Department of Chemistry
Iowa State University
1605 Gilman Hall
Ames, IA 50011
Tel.: +1 515-294-8356
E-mail address: andersoj@iastate.edu
ORCID: 0000-0001-6915-8752

Keywords: Fish; Personal care products; Matrix solid-phase dispersion; Solid-phase microextraction; GC-MS.

1. Introduction

Emerging contaminants (ECs) are compounds whose existence raises concern regarding their toxicity, despite the fact they are not yet regulated by regulatory agencies, as there is little information about their concentration levels in the environment (Sauvé & Desrosiers, 2014). Among ECs, personal care products (PCPs) comprise a wide range of compounds and include UV filters (UVFs), synthetic musk fragrances, insect repellents, antimicrobials, and preservatives (Buchberger, 2011). These compounds are commonly used as ingredients in a broad range of daily products for hygiene and skin care (e.g., deodorants, lotions, toothpaste, cosmetics, perfumes, shampoos, detergents, soaps, sunscreens, and insect repellents, among others). They are currently of concern due to their extensive use, ubiquity in the environment, pseudo-persistence, and potential toxicity (Montesdeoca-Esponda et al., 2018). Many PCPs, such as parabens, UV filters, polycyclic musks, and antimicrobials, are already designated as endocrine disrupting compounds (Witorsch & Thomas, 2010). UVFs are used in sunscreens to protect the skin against UVA (315-400 nm) and UVB (280-315 nm) radiation. These compounds typically contain aromatic moieties with methoxy or carbonyl substituent groups that provide high absorbance of UV light (Hopkins & Blaney, 2016), with the two most common examples being avobenzone and octocrylene (Jesus et al., 2022). Synthetic musk fragrances are compounds added as scents in various products used for personal care, with polycyclic musks galaxolide and tonalide being the most prominent. The most common insect repellent is N,N-diethyl-meta-toluamide and it is employed to protect against mosquitoes and biting flies. Antimicrobials assist in preventing the growth of unwanted microorganisms and extend the expiration of various products. Triclosan, triclocarban, and chlorophene are commonly employed as antibacterial agents. Finally, preservatives are also employed to prevent the growth of microorganisms. Parabens are p-hydroxybenzoic acid

derivatives and represent the largest fraction of the preservative market, with methyl and propylparaben being among the most frequently employed in personal hygiene products and cosmetics (Hopkins & Blaney, 2016).

PCPs have been frequently detected in aquatic organisms, including various invertebrates and fish, at levels down to the ng to μg per gram of tissue, (Ramirez et al., 2009) indicative that these compounds can be bioaccumulated and biomagnified through the food chain (Chen et al., 2017). During the production chain of fish, which includes growth, slaughter, processing to produce fish fillets, and the distribution of fish products to the market, potential danger exists in contamination with PCPs. Fish may absorb the PCPs from water as well as when they eat contaminated feed. PCPs may also be introduced through the handling of fish products in the manufacturing process, including packaging and distribution (Eguiraun et al., 2015). While most studies that determine PCPs in fish have focused on a single group of compounds (Núñez et al., 2017), PCPs are present in the environment within complex mixtures. The simultaneous monitoring and detection of the various PCP groups is challenging due to their diverse physicochemical properties as well as their presence at low concentration levels, which can be further complicated by the presence of numerous potentially interfering matrix components, such as lipids. In addition, the fat content and fatty acid composition of fish lipids are extremely variable, even within species, and are dependent upon different factors such as the season of the year, type and amount of feed, reproductive cycle, as well as the temperature, pH, and salinity of the water. For example, the lipid content can vary from 4 to > 30 % (w/w) in mackerel, depending on the season (Moradi et al., 2011). For this reason, Mottaleb et al. (2009) developed two analytical methods employing different conditions that depend on the lipid content of the matrix. For bluegill (muscle tissue ~0.4% w/w of lipids), a method based on solid-liquid extraction followed by GC-

MS was found to be appropriate, but it was unsuitable in the analysis of tissues containing >1% (w/w) of lipids. In contrast, Sonora sucker (muscle ~4.9% w/w of lipids) required another method using solid-liquid extraction followed by gel permeation chromatography (GPC) clean-up to remove lipids and was coupled to GC-MS/MS to improve sensitivity.

To overcome the aforementioned challenges, selection of the appropriate sample preparation technique is a critical step to ensure removal of matrix interferences while enriching analytes to detectable concentration levels. Moreover, one or more clean-up steps is nearly unavoidable due to the high lipid content of fish (Núñez et al., 2017). Most current methods used to extract PCPs from fish are based on the partitioning of analytes between the sample matrix and an organic solvent. Soxhlet extraction (Meinerling & Daniels, 2006), ultrasound-assisted extraction (UAE) (Peng et al., 2015), pressurized liquid extraction (PLE) (Kim et al., 2011), Quick, Easy, Cheap, Effective, Rugged, and Safe (QuEChERS) (Jakimska et al., 2013) and, to a lesser extent compared to the above mentioned, matrix solid-phase dispersion (MSPD) (Tsai et al., 2014; Xiao et al., 2021) have been reported. After extraction, an additional clean-up step, such as solid-phase extraction (SPE) or GPC, is required to remove co-extracted compounds that can often interfere with chromatographic analysis. Deterioration of chromatographic performance is evidenced by substantial shifts in analyte retention times, tailing or fronting of the peaks, increased background signal, and reduction in analyte sensitivity (Mottaleb et al., 2009; Núñez et al., 2017). The excessive time required for extraction, requirements of large sample amounts, high solvent consumption, in addition to limited selectivity, are among some of the important drawbacks of these approaches. Approaches that result in high selectivity and sensitivity are required due to the low concentration of PCPs in samples. The use of liquid chromatography-mass spectrometry (LC-MS) or GC-MS are commonly employed for trace level analysis. In recent years, there has been a

movement in analytical chemistry to reduce sample and solvent consumption by miniaturizing the involved procedures. The movement has led to the emergence of various microextraction techniques including solid-phase microextraction (SPME), stir bar sorptive extraction (SBSE), dispersive solid-phase extraction (d-SPE), and numerous techniques belonging to liquid-phase microextraction (LPME) (Wianowska et al., 2020). The simplicity of MSPD and its combination with SPME, a solvent-free sample preparation technique, makes them an ideal combination for developing miniaturized analytical methods and are more attractive in the context of Green Analytical Chemistry.

MSPD is an approach that mechanically blends the solid sample with a solid support material resulting in complete sample disruption and enhanced analyte isolation. The resulting mixture can be transferred and packed into an empty cartridge, following by the elution of analytes using an appropriate solvent (Barker, 2007). Miniaturization of MSPD (sometimes called micro-MSPD) can be achieved by reducing the amount of sample from the typical gram to milligram level to decrease the consumption of solid support and organic solvent, as well as shorten analysis times (Kristenson et al., 2006). Some disadvantages of MSPD are that it is a fairly labor-intensive technique, the ratio between the sample and the eluent results in dilution of the analytes in the extract, a reduction in sensitivity, and lack of automation for the procedure. SPME, on the other hand, involves a fiber coated with a stationary phase that is exposed to an aqueous sample until equilibrium is established between the analyte in the sample and on the fiber. Analytes are then desorbed from the fiber at high temperatures in a GC injector, completely eliminating the use of organic solvents, and having advantages of being simple, rapid, yielding high sensitivity, and it can be automated (Kataoka et al., 2000) which can compensate for the disadvantages of MSPD. Although MSPD and SPME are not new extraction methods, very little has been reported regarding

their combined application for the efficient extraction of organic contaminants from biota samples. Their combination has been demonstrated in the analysis of persistent organic contaminants in aquatic organisms, such as polycyclic aromatic hydrocarbons (Campíns-Falcó et al., 2008), pesticides, and polybrominated diphenylethers (Moliner-Martinez et al., 2009) in bivalves. There have been no studies regarding their use in the extraction of ECs.

The objective of this work was to develop a sensitive, selective, and simple miniaturized MSPD-SPME-GC-MS method to simultaneously determine eleven PCPs in fish samples avoiding the use of special equipment or derivatization of the analytes. The target analytes were from the following five main groups of PCPs: musks, UV filters, preservatives, antimicrobials, and insect repellents. The performance of the method was assessed with samples of tilapia and salmon, fish containing low and high lipid content, respectively. The method was also applied in the analysis of eight commercial fish samples, including four tilapia and four salmon, obtained from local supermarkets.

2. Experimental

2.1. Chemicals, reagents, materials and samples

The PCPs studied and determined in this work included two antimicrobials, two musks, four UV filters, two preservatives, and one insect repellent. The analytes N,N-diethyl-meta-toluamide (DEET, 98.5%), chlorophene (CP, >95%), tonalide (TON, >97.0%), methyl paraben (MP, 100.0 %, analytical standard), propyl paraben (PP, 100.0 %, pharmaceutical secondary standard), oxybenzone (OXY, >97.5 %), and 3-(4-methylbenzylidene)camphor (4-MBC, >98.0%) were purchased from Sigma-Aldrich (St. Louis, MO, USA). The analytes triclosan (TCS, >98.0%), octocrylene (OCT, >98.0%), 2-ethylhexyl 4-(dimethylamino)benzoate (EHPABA, >98.0%), and

154 galaxolide (GAL, 50% in diethyl phthalate) were purchased from Tokyo Chemical Industry (TCI,
155 Chuo-ku, Tokyo, Japan). Internal standards (ISs) of anthracene (ANT, 99%) and
156 benz[a]anthracene (B[a]A, 99%) were also obtained from Sigma-Aldrich. Individual stock
157 solutions of the analytes and ISs were prepared in acetonitrile (ACN, > 99.9 %, Sigma-Aldrich) at
158 a concentration of 2000 mg L^{-1}. Intermediate solutions containing all of the analytes were prepared
159 in acetonitrile by dilution of the individual stock solutions. Working solutions were prepared by
160 spiking appropriate amounts of the intermediate solutions into the sample or ultrapure water,
161 depending on the experiment. All solutions were stored in glass vials at 4 °C and were covered to
162 protect them from light.

163 Ultrapure water (18.2 MΩ·cm resistivity) was obtained from a Milli-Q water purification
164 system (Millipore, Bedford, MA, USA). Sodium chloride (NaCl, ≥99.5%) was purchased from
165 Fisher Scientific (Fair Lawn, NJ, USA). Dichloromethane (≥99.5%) and hexane (≥98.5%) were
166 obtained from Sigma-Aldrich. Discovery® DSC C18 (particle diameter 50 μm) sorbent was
167 obtained from Sigma-Aldrich. Bondesil PPL (particle diameter 125 μm), Bond Elut Enhanced
168 Matrix Removal (EMR)-lipid, Bond Elut EMR-lipid polish (NaCl/anhydrous $MgSO_4$ (4:1, w/w),
169 polypropylene syringes (6 mL capacity) and 20 μm polyethylene frits were provided as gifts from
170 Agilent Technologies (Santa Clara, CA, USA). An agate mortar (100 mm O.D. x 82 mm I.D. x 25
171 mm depth) with a pestle was acquired from MSE supplies (Tucson, AZ, USA).

172 Commercial SPME fibers consisting of polyacrylate (PA, 85 μm of film thickness),
173 carboxen/polydimethylsiloxane (CAR/PDMS, 75 μm of film thickness),
174 divinylbenzene/carboxen/polydimethylsiloxane (DVB/CAR/PDMS, 50/30 μm of film thickness),
175 polydimethylsiloxane/divinylbenzene (PDMS/DVB, 65 μm of film thickness) and carbowax-

176 polyethylene glycol (PEG, 60 μm of film thickness) sorbent coatings were supplied by Sigma-
177 Aldrich. Commercial SPME Arrows comprised of divinylbenzene/polydimethylsiloxane
178 (DVB/PDMS, 120 μm of film thickness, 1.5 mm needle diameter) and divinylbenzene/carbon
179 wide range/polydimethylsiloxane (DVB/Carbon WR/PDMS, 120 μm of film thickness, 1.5 mm
180 needle diameter) sorbents were provided as gifts from Restek (Bellefonte, PA, USA). All fish
181 samples were purchased at supermarkets in Ames, Iowa, USA. Tilapia and salmon muscle were
182 homogenized in a food processor (Mainstays, Guangdong, China) and freeze dried using a
183 lyophilizer (Labconco, Kansas City, MO, USA).

184 **2.2. Instrumentation**

185 Analyses were carried out using a 7890B GC system equipped with 5977A MS detector
186 (single quadrupole) from Agilent Technologies (Santa Clara, CA, USA). A Rtx-5ms capillary
187 column (30 m × 0.25 mm I.D. × 25 μm film thickness) from Restek (Bellefonte, PA, USA) was
188 used for the separation of analytes. Ultrapure helium was used as carrier gas at a flow rate of 1 mL
189 min^{-1}. The inlet was operated in *splitless* mode for 1 min with a temperature of 270 °C. The
190 following oven temperature program was used to separate the analytes: initial temperature of 120
191 °C during 1 min, then the temperature was increased at 10 °C min^{-1} to 300 °C, and held for 5 min.
192 The transfer line from the GC to the MS was kept at 280 °C. The MS was operated in electron
193 ionization (EI) mode at 70 eV. The ion source and quadrupole temperatures were 230 °C and 150
194 °C, respectively. Data were acquired in single ion monitoring (SIM) mode. For analyte
195 identification, the following three identification parameters were considered: retention time, the
196 presence of three characteristic ions of each analyte, and their ratio. The ions monitored for each
197 analyte and the internal standards were as follows (quantifier ion in bold): MP 93, **121**, 152 m/z;

198 DEET 91, **119**, 190 m/z; PP **121**, 138, 180; ANT (IS) 76, 152, **178** m/z; GAL and TON 159, 213,
199 **243**, 258 m/z; CP 140, 183, **218** m/z; OXY 77, **151**, 228 m/z; 4-MBC 105, 128, **254** m/z; TCS 146,
200 218, **288** m/z; EHPABA 148, **165**, 277 m/z; B[a]A (IS) 114, 226, **228** m/z; and OCT 204, **249**, 360
201 m/z. All data were acquired using Mass Hunter Workstation software from Agilent Technologies
202 version B.07.00. For quantitative purposes, the peak area of the quantifier ion was employed. Table
203 S2 of the Supplementary Information shows the retention times for each analyte.

204 **2.3. Solid-phase microextraction (SPME)**

205 For optimization of the SPME method, the analytes were spiked in water at 100 ng mL^{-1}.
206 The following parameters were evaluated using a one-factor-at-a-time approach: type of fiber (PA,
207 CAR/PDMS, DVB/CAR/PDMS, PDMS/DVB, and PEG), organic solvent (2, 5, 9 % ACN, v/v),
208 ionic strength (0, 10, 18 % NaCl, w/v), temperature (25, 35, 45, 55 °C), extraction time (15, 30,
209 45, 60 min), and injector desorption temperature (250 and 270 °C). Other parameters including the
210 use of direct-immersion mode, volume of diluted sample (15 mL) and stirring rate (1000 rpm)
211 were fixed. Direct-immersion mode was selected because most of the analytes were of low to
212 medium volatility and high to medium polarity (Table S1). All experiments were performed in
213 triplicate.

214 **2.4. Matrix solid-phase dispersion (MSPD)**

215 To optimize the MSPD method, fish samples spiked at 500 µg g^{-1} were used and the extracts
216 analyzed with the optimum conditions of the SPME-GC-MS method, unless otherwise specified.
217 The following parameters were evaluated using a one-factor-at-a-time approach: type of dispersant
218 sorbent (Discovery® DSC C18 and Bondesil PPL), ratio sample-sorbent (1:2, 1:4, 1:6, 1:8, 1:10),
219 clean-up (EMR-lipid, EMR-lipid + EMR lipid polish, EMR-lipid as co-column), elution solvent

(ACN, ACN-water 80:20, 85:15 and 90:10 (v/v) ratios) and elution volume (1, 2, 3, 4, and 5 mL). All experiments were performed in triplicate.

2.5. Optimum conditions

The optimum conditions for MSPD were as follows: 25 mg of lyophilized muscle and 150 mg of Bondesil PPL sorbent were blended in an agate mortar until a homogeneous mixture was obtained. The mixture was then placed into a 6 mL polypropylene cartridge containing 300 mg of Bond Elut EMR-lipid as co-column and clean-up sorbent. Analytes were eluted from the cartridge with 3 mL of acetonitrile-water 90:10 (v/v).

The optimum conditions for SPME were as follows: 1 mL of the MSPD extract was diluted with 14 mL of deionized water. The solution was transferred into a 20 mL headspace vial (Restek), with the addition of 20 μL of the IS solution (Ant and B[a]A at 50 μg mL^{-1} in ACN) and 1.5 g of NaCl (10 % w/v). The solution was agitated by vortex for 1 min. A stir bar (1 cm length × 0.5 cm diameter, from Fisher Scientific) was added, and the vial was sealed using a polytetrafluoroethylene (PTFE) crimp cap (Restek). The vial was placed on a Corning PC-420D magnetic stirring hotplate (Corning, NY, USA), and DI-SPME was performed by inserting the fiber in the solution for 45 min at 35 °C while fixing the stir rate to 1000 rpm. After extraction, the fiber was rinsed in water before desorption. Thermal desorption was performed in the GC injector for 1 min at 270 °C in splitless mode, and the fiber was allowed to remain in the inlet for 15 min to prepare the fiber for the next sample and avoid carryover effects.

A summary of the complete MSPD-SPME-GC-MS method with the optimum conditions is shown in Figure 1.

2.6. Method validation

Once the MSPD-SPME-GC-MS method was optimized, analytical parameters including matrix effects, accuracy, precision (repeatability), linearity, limit of detection (LOD), and limit of quantitation (LOQ) were evaluated for tilapia and salmon. Matrix effects were evaluated by comparing the average area ratios of standards containing all analytes in the pure solvent at 56 ng mL^{-1} and matrix-matched standards at 56 ng mL^{-1} (equivalent to 100 μg g^{-1}) for the two samples, with each standard being prepared in triplicate. Accuracy and precision were assessed by analyzing blank samples spiked at four concentration levels of 5, 10, 100 and 300 μg g^{-1}. For each concentration, three replicates were made, and the recoveries and relative standard deviations (RSDs) were calculated for each level. LOD and LOQ were determined with a signal-to-noise ratio (S/N) of 3 and 10, respectively, using matrix-matched standards (tilapia and salmon) prepared in duplicate from 0.05 to 150 ng mL^{-1}. Linearity of the method was evaluated with matrix-matched standards (tilapia and salmon) at five concentration levels for MP (150, 200, 300, 400, and 500 μg g^{-1}) and seven concentration levels (10, 50, 100, 200, 300, 400, and 500 μg g^{-1}) for the remaining analytes. For each concentration level, three replicates were made. The coefficient of determination (R^2), slope and Y intercept of the calibration curves were calculated.

3. Results and discussion

3.1. Optimization of the SPME procedure

For optimization of the SPME method, which was used to analyze the MSPD extracts (Figure 1), the following most influential parameters were studied: type of fiber, organic solvent content, ionic strength, extraction temperature, extraction time, and desorption temperature. Other parameters, including sample volume and stirring rate, were fixed. The sample volume was fixed

263 at 15 mL to ensure suitable preconcentration by the SPME fiber. Moreover, direct-immersion
264 mode was employed with a high stirring rate (1000 rpm). Mass transfer of the analytes from the
265 sample to the extracting phase is limited by the diffusion of the analytes through the static layer
266 surrounding the fiber, called the Prandtl boundary. The thickness of the boundary layer largely
267 depends on the degree of agitation; therefore, high stirring rates shorten the equilibration time
268 (Pawliszyn, 2012).

269 The main challenge of the SPME method was to extract the eleven PCPs simultaneously
270 despite their different chemical structures and wide ranging physicochemical properties (Table
271 S1). For this reason, screening of the commercial SPME fibers with various sorbent coatings and
272 polarities, including PA, CAR/PDMS, DVB/CAR/PDMS, PDMS/DVB, and PEG, was performed.
273 The PDMS fiber is generally preferable for hydrophobic analytes, while PA and PEG fibers are
274 for more polar compounds (Kataoka et al., 2000). Figure S1 shows the extraction efficiencies for
275 each analyte using all fibers, expressed as chromatographic peak areas. All fibers showed low
276 extraction efficiencies for methyl paraben, a highly polar and non-volatile compound, and
277 octocrylene, the most hydrophobic compound. Using the PA fiber, the peak of DEET was not
278 detected, while CAR/PDMS, DVB/CAR/PDMS, and PEG fibers exhibited poor extraction for the
279 majority of analytes. The results clearly showed that PDMS/DVB was the most suitable fiber due
280 to its extraction efficiency being the highest for most analytes. Other reported SPME methods for
281 the determination of PCPs in water commonly use the PDMS fiber for UV filters and musks due
282 to their non-polar character, and the DVB/CAR/PDMS fiber for parabens and antimicrobials
283 (Portillo-Castillo et al., 2018). In this study, PDMS/DVB was found to be the most suitable, as the
284 amount of analyte extracted by the fiber from a water sample can be significantly affected by the
285 type of fiber and the kind of analytes present in the sample matrix.

In aqueous matrices, the content of organic solvent should be between 1 and 5% (v/v) to avoid influencing partitioning of analytes to the fiber. The distribution constant of the analyte between the sample and fiber is expected to decrease substantially when the polarity of the aqueous sample decreases and the coating is swollen by the organic solvent present in the matrix (Pawliszyn, 2012). However, when dealing with the extraction of complex matrices such as fish, adding organic solvent can improve analyte recovery by altering the binding interactions of the co-extracted matrix compounds (such as lipids) with the analytes, thereby promoting the free form of analytes (Gionfriddo et al., 2020). Acetonitrile was added at levels from 2 to 9 % (v/v) to evaluate the effect of organic modifier as well as avoid evaporation of the MSPD extract. As shown in Figure S2, the highest extraction efficiency was achieved at an acetonitrile content of 2 % (v/v) for the most polar compounds (MP, PP, DEET). When the amount of organic solvent is increased, analytes are prone to be dissolved in the organic solvent and not be extracted by the fiber. The opposite effect was observed for the most hydrophobic analytes (EHPABA and OCT) with the highest extraction efficiency being attained using an acetonitrile content of 9 % (v/v), as the addition of organic solvent increases their solubility and prevents their adsorption on the inner surface of the glass vial. On the other hand, similar extraction efficiency was obtained with an acetonitrile content of 2 % and 6 % (v/v) for analytes having intermediate polarity (GAL, TON, CP, OXY, 4-MBC, and TCS), while a significant decrease was observed at 9 % (v/v). Based on the results, an acetonitrile content of 6 % (v/v) was selected as it offered the highest extraction efficiency and good precision for most of the analytes.

Enhancing the ionic strength by the addition of salt is known to increase the extraction of analytes through the salting-out effect (Kataoka et al., 2000). This effect is related to the fact that adding salt to the sample decreases analyte solubility and increases their mass transfer to the fiber

309 (Pawliszyn, 2012). In this study, the salting-out effect was tested by adding sodium chloride at
310 concentrations ranging from 0 to 18 % (w/v). According to Figure S3, the addition of salt and the
311 ensuing change in ionic strength significantly enhanced the extraction efficiency for the most polar
312 analytes (MP, DEET, PP), as the salting-out effect is more pronounced with increasing compound
313 polarity (Pawliszyn, 2012). On the other hand, for the mid-polar and non-polar analytes, the
314 addition of salt did not provide higher recovery. The best extraction efficiency was obtained
315 without salt as the presence of salt increased the viscosity of the sample, slowing the diffusion of
316 the analytes to the fiber (Canosa et al., 2006). In order to enhance the extraction efficiency of polar
317 analytes, which also exhibit the lowest responses, an intermediate concentration of 10 % (w/v) was
318 used for subsequent studies.

319 Temperature plays a significant role in the sensitivity of SPME as it affects the extraction
320 kinetics. An increase in extraction temperature results in an increase in extraction rate and
321 simultaneously decreases the distribution constant of analyte between the matrix and the fiber
322 (Pawliszyn, 2012). Extractions were studied in a range from 25 to 55 °C, and the behavior of the
323 analytes followed two different patterns. Figure S4 shows that for most of the compounds, an
324 enhancement in extraction efficiency was observed at a temperature up to 55 °C, but method
325 repeatability was compromised, mainly for the semi-volatile musks. The opposite effect was
326 observed for parabens, as their extraction efficiencies were similar at 25 °C and 35 °C, but then
327 decreased when the temperature was higher than 45 °C. To establish the best compromise in terms
328 of analyte response, an intermediate temperature of 35 °C was selected.

329 Extraction time is another important parameter to consider as SPME is not an exhaustive
330 extraction technique, and consists of a multi-phase equilibration process where the maximum
331 extraction efficiency is achieved under equilibrium conditions. Determination of the optimum

extraction time is required to achieve the desired sensitivity. Extraction-time profiles were carried out from 15 to 60 min to determine the optimal extraction time for further method development. These profiles revealed that the amount of analytes extracted increases rapidly in the first 45 min, followed by a smaller increase from 45 min to 60 min. The majority of compounds did not attain equilibration within the experimental conditions tested, and only MP was found to reach equilibrium at 45 min (Figure 2). However, full equilibration is not necessary in SPME for accurate analysis due to the linear relationship between the amount of analyte adsorbed by the fiber coating and its initial concentration in the sample matrix under pre-equilibrium conditions (Kataoka et al., 2000). In order to establish the best compromise between analyte response and analysis throughput, an extraction time of 45 min was selected.

Desorption temperature plays an important role in accelerating the transfer of analytes from the SPME fiber to the chromatographic column. Typically, SPME fibers should be desorbed at high temperatures to ensure efficient desorption and rapid release of analytes from the injector. For the PDMS/DVB fiber, desorption temperatures of 250 and 270 °C were tested since the majority of analytes possess medium volatility. When a temperature of 270 °C was employed, the peak areas for all analytes were higher compared with those at 250 °C (Figure S5). A 100% increase was observed for the more polar (MP, DEET, and PP) and the more hydrophobic (EHPABA and OCT) analytes, which also exhibited the lowest response. To increase method sensitivity, an optimum desorption temperature of 270 °C was selected.

3.2. Optimization of the MSPD procedure

Among the main drawbacks of chromatographically-determining organic contaminants in fish is that lipids are commonly co-extracted with the analytes and can contaminate the GC-MS

354 system. Lipids are known to worsen the shape of chromatographic peaks, shorten the useful life of
355 capillary columns, and contaminate the MS ion source. Methods reported in the literature are
356 usually developed for a specific kind of matrix due to the variability in the lipid content of fish.
357 For this reason, the MSPD method featured in this study for the extraction of analytes from fish
358 muscle (Figure 1) was optimized considering its application to a wide range of fish matrices.
359 Tilapia and salmon were selected because their lipidic content was approximately 4 % and 40 %
360 (w/w), respectively (Table S3). The MSPD conditions were optimized using acetonitrile as the
361 elution solvent because of its low affinity for lipids (Negreira et al., 2013). The following four
362 parameters were studied in the MSPD extraction: type of dispersant sorbent, sample-sorbent ratio,
363 clean-up, and elution volume.

364 Sorbent selection is of utmost importance since it is one of the variables controlling the
365 selectivity of the MSPD process. The chemical composition and characteristics of the solid sorbent
366 and bonded phase are expected to affect the retention and elution of analytes. The bonded phase
367 acts like a solvent that dissolves and disperses sample components onto the sorbent surface based
368 on their relative polarities, consequently achieving complete disruption of the sample. As an
369 example for the C18 sorbent, hydrophobic matrix components are dispersed within the non-polar
370 organic bonded phase, with polar molecules capable of forming hydrogen bonding (like water)
371 being associated with the free silanols of the silica particle and distribution of larger molecules
372 across the surface of the multi-phasic structure (Barker, 2000; Barker, 2007). Most MSPD
373 applications employ silica-based sorbents, such as C18 and inorganic sorbents (silica, florisil, and
374 alumina), but polymeric sorbents have been under studied (Capriotti et al., 2010). For this reason,
375 the silica-based sorbent Discovery® DSC C18 and the polymeric sorbent Bondesil PPL were
376 tested in this work. To select the optimum sorbent, 50 mg of tilapia sample was dispersed with 200

377 mg of sorbent (Discovery® DSC C18 or Bondesil PPL) and eluted with 5 mL of acetonitrile. One
378 microliter of the extract was injected into the GC-MS system. The chromatograms obtained in scan
379 mode for the MSPD extracts using the C18 and Bondesil PPL sorbents are shown in Figure S6.
380 Peaks of three unknown matrix compounds were detected in both extracts. However, the height
381 of the peaks was two times more (indicating a higher concentration) in the C18 extract than in the
382 Bondesil PPL. Also, from 18 min to 18.5 min, two additional peaks of unknown matrix
383 components were detected only in the chromatogram from the C18 extract. Based on the results,
384 the polymeric Bondesil PPL sorbent was selected for subsequent optimization studies, as fewer
385 matrix components were co-extracted with the analytes. This may be due to the chemical
386 composition of the Bondesil PPL sorbent, which consists of a proprietary functionalized styrene
387 divinylbenzene polymer, that unlike the silica-based sorbent C18, provides chemical selectivity
388 for aromatic hydrophobic and certain polar compounds such as phenols (Li et al., 2016).

389 The sample-to-sorbent ratio is important because it exposes the surface of the sample to
390 solvent during the elution step (Figure 1). For most studies, sample-to-sorbent ratios range from
391 1:1 to 1:4, but higher ratios have been also applied depending on the application (Capriotti et al.,
392 2010). Several sample-to-sorbent ratios were tested for tilapia and salmon (Table S4). As shown
393 in Figure S7, dilution of tilapia extracts with water obtained for ratios of 1:2 and 1:4 resulted in a
394 non-homogeneous solution, while the 1:6 ratio provided a transparent solution. On the other hand,
395 dilution of the salmon extracts with all of the ratios studied resulted in a non-homogeneous solution
396 with the presence of a white solid, as shown in Figure S8. The 1:6 ratio was selected for subsequent
397 experiments because no further improvement was observed when the ratio was increased.

398 A clean-up step with an appropriate sorbent to remove co-extracted matrix compounds was
399 necessary to obtain a transparent diluted extract suitable for DI-SPME. Typically, clean-up of the

MSPD extract is performed with solid phase extraction (SPE) (Capriotti et al., 2010). To evaluate the clean-up step, the EMR-lipid sorbent was selected because it was demonstrated previously to selectively and efficiently remove lipids in the QuEChERS extraction of polycyclic aromatic hydrocarbons (Urban & Lesueur, 2017) and phenolic compounds (Yin et al., 2022) from salmon. The following three approaches were tested as clean-up: (1) dispersive-SPE (d-SPE) with 200 mg of EMR-lipid sorbent, (2) d-SPE with 200 mg of EMR-lipid sorbent followed by d-SPE with 200 mg of EMR-lipid polish (a post d-SPE step that improves the removal of water and solid residues from the sample extract), and (3) 300 mg of EMR-lipid sorbent as a co-column packed into the bottom of the same cartridge as the MSPD material. The use of EMR-lipid sorbent as co-column was selected since it was found to produce transparent diluted extracts for the tilapia and salmon matrices. However, the scan chromatogram obtained for tilapia after SPME revealed that lipids were still co-extracted (Figure S9). To address this, two modifications of the method were performed in subsequent experiments. First, the sample amount was decreased while maintaining a sample-to-sorbent ratio of 1:6 (25 mg of sample to 150 mg of PPL sorbent). Second, the strength of the elution solvent was decreased to enhance performance of the EMR-lipid sorbent, as it is highly recommended to perform the extraction with organic solvent-water 80:20 (v/v) for optimal matrix removal (Agilent Technologies, 2017). Therefore, different ACN-water ratios (80:20, 85:15, and 90:10 (v/v)) were tested for salmon MSPD extractions. Scan chromatograms obtained from these extracts are shown in Figure S10. A significant enhancement of lipid removal was observed with the three tested eluents compared to when using only acetonitrile as the eluent solvent (Figure S9). The lowest intensity of the lipid peak (observed from 15 to 17 min) was obtained using the 80:20 (v/v) ratio, while a similar intensity was seen for 85:15 and 90:10 (v/v) ratios. For this reason, the elution profile was evaluated with the 80:20 and the 90:10 (v/v) ratios.

The elution profile of analytes from the MSPD cartridge was obtained to determine the smallest volume of solvent (ACN-water 80:20 and 90:10 ratios) required to elute all of the target analytes. The elution profile was recorded by fractionated elution of the MSPD cartridge for tilapia and salmon samples, with a volume of each fraction of 1 mL and a total elution volume of 5 mL. Each fraction was analyzed individually, and the accumulated recovery was calculated by comparing the amount of analytes obtained in each fraction with the total amount of analytes initially added. Results for the ACN-water 80:20 (v/v) ratio are shown in Figure S11. For the polar analytes (MP, PP and DEET) with 5 mL of eluent, the accumulated recovery was > 95 % and > 114 % for tilapia and salmon, respectively. For the more hydrophobic analytes (EHPABA and OCT) with 5 mL of eluent, the accumulated recovery was around 40 % and 60 % for tilapia and salmon, respectively. Results for the ACN-water 90:10 (v/v) ratio are shown in Figure S12. When a 3 mL volume was eluted, the accumulated recovery was > 90 % for all analytes. The results were similar for both matrices due to increased strength of the eluent. For this reason, ACN-water 90:10 (v/v) was selected as eluent.

3.3. MSPD-SPME-GC-MS method validation

The MSPD-SPME-GC-MS method developed in this study was validated in terms of matrix effect, precision (repeatability), accuracy, linearity, LOD, and LOQ. Matrix effects were evaluated by comparing of the average relative area of the analytes in pure acetonitrile and matrix-matched standards (as described in section 2.4.1), and the results are presented in Figure S13. For both fish samples, a significant difference between the average relative areas was found for most of the analytes using a 95% confidence level. Polar analytes (MP, DEET, and PP) showed a positive matrix effect, while musks (GAL and TON) and EHPABA exhibited a negative matrix effect. Additionally, CP and OXY also showed a positive matrix effect for the spiked salmon

446 extracts as the matrix contained higher amounts of lipid compared to tilapia. These results indicate
447 that matrix effects for these samples could not be fully compensated by the use of internal standards
448 for all analytes. For this reason, matrix-matched calibration was employed to enable quantitative
449 analysis.

450 Results of the validation parameters for tilapia and salmon are shown in Tables 1, 2 and 3.
451 Precision was evaluated in terms of repeatability, and results for tilapia and salmon are presented
452 in Tables 1 and 2, respectively. For 100 μg g^{-1}, RSDs ranged from 2.83 % to 12.04 % for tilapia,
453 and from 3.30 % to 12.96 % for salmon. For 300 μg g^{-1}, RSDs ranged from 2.59 % to 10.92 % for
454 tilapia, and from 1.62 % to 8.22 % for salmon. Regarding the low spiked levels of 5 and 10 μg g^{-1},
455 the RSD values were < 11.82%. The results showed that acceptable precision could be obtained
456 for both matrices with RSD < 13% for all analytes.

457 Accuracy was evaluated at the same concentration levels used for precision studies; for 100
458 μg g^{-1}, the recoveries ranged from 85.79 % to 108.63 % for tilapia (Table 1), and from 83.13 % to
459 105.98 % for salmon (Table 2). For 300 μg g^{-1}, the recoveries ranged from 80.11 % to 99.91 % for
460 tilapia and from 82.19 % to 107.39 % for salmon. Regarding the low spiked levels of 5 and 10 μg
461 g^{-1}, the recoveries were > 90% for all the analytes in salmon and tilapia, except for MP which had
462 higher LOQ values. According to the results, acceptable accuracy was obtained for both matrices
463 with recoveries > 80 % for all analytes.

464 Adequate linearity was obtained for both samples, with coefficients of determination
465 ranging from 0.97 to 0.99 and acceptable standard error for the slope and Y intercept (Araujo,
466 2009) (Tables 1 and 2). LODs and LOQs were considered with the dry (freeze-dried tissue) and
467 wet (fresh tissue) weight of the samples and the results are presented in Table 3. For the dry weight,

LODs ranged from 0.04 μg g^{-1} to 44.95 μg g^{-1} for tilapia and from 0.04 μg g^{-1} to 39.23 μg g^{-1} for salmon while LOQs ranged from 0.12 μg g^{-1} to 149.83 μg g^{-1} for tilapia and from 0.13 μg g^{-1} to 130.77 μg g^{-1} for salmon. The LODs were higher for salmon than for tilapia due to fewer co-extracted compounds from the matrix. Considering that wet weight represents the fresh samples as they are consumed, LODs ranged from 0.01 μg g^{-1} to 10.46 μg g^{-1} for tilapia and from 0.01 μg g^{-1} to 14.36 μg g^{-1} for salmon. The results demonstrate the method's high sensitivity which uses only 25 mg of dry sample and avoids analyte derivatization. However, for some of the analytes such as MP, PP, DEET, and OCT, the obtained LODs surpassed the ng g^{-1} level reported in previous studies (Kim et al., 2011; Mottaleb et al., 2009). To overcome this challenge, SPME Arrow was investigated to increase the extraction of analytes in an effort to enhance the method's overall sensitivity.

3.4. Enhancing method sensitivity using SPME Arrow

SPME Arrow was recently introduced as an alternative extraction technique to overcome some drawbacks associated with traditional SPME, including the limited mechanical robustness of the fiber and the small sorption phase volume of the commercially-available fibers. SPME Arrow combines a larger sorbent volume with high mechanical robustness, providing a higher extraction capacity than traditional SPME to achieve trace-level sensitivity. Contrary to classical SPME fibers, the SPME Arrow design fully shields the sorbent material, reducing risk of contamination from ambient air and loss of analytes during transfer processes. SPME Arrow is also compatible with desorption in a standard GC injection port due to its dimensions and sharp tip (Herrington et al., 2020). Most of the reported methods that have used SPME Arrow are in headspace mode, and it has been employed to determine synthetic musk fragrances in fish samples (Castro et al., 2019).

SPME Arrow was investigated in this work to increase the sensitivity of the MSPD-SPME-GC-MS method (Figure 1). The experiments were performed with water spiked at 100 ng mL^{-1}. The DVB/PDMS and DVB/Carbon WR/PDMS arrows were tested in direct-immersion mode using the following conditions: sample volume 15 mL, extraction temperature 35 ºC, ACN 5 % (v/v), NaCl 10 % (w/v), stirring rate 1000 rpm, and extraction time 3 h. After extraction, thermal desorption was performed in the GC injection port for 1 min at 270 °C in splitless mode, with the fiber remaining in the inlet for 15 min to avoid carryover effects. The content of ACN was decreased to 5 % (v/v) to avoid swelling of the fiber coating.

As shown in Figure S14, the DVB/Carbon WR/PDMS arrow provided higher analyte responses than the DVB/PDMS arrow, with the exception of EHPABA and OCT. MP was also not detected using the DVB/PDMS arrow. The DVB/Carbon WR/PDMS arrow was also tested with matrix-matched standards spiked at 100 ng mL^{-1} and containing 0.5 mL of MSPD of fish extract. As observed in Figure S15, some lipid peaks were detected between 13 and 15 min in the scan chromatogram for the tilapia and salmon samples. As expected, the peak intensities were higher in the salmon sample. However, the lipid peaks did not interfere with analyte peaks in the SIM mode, as seen in Figure S16, indicating that both fish samples were suitable for analysis with SPME Arrow.

LODs and LOQs were calculated with matrix-matched standards for tilapia and salmon, based on a signal-to-noise ratio of 3 and 10, respectively. Considering the dry weight, LODs ranged from 0.004 μg g^{-1} to 1.977 μg g^{-1} for tilapia and from 0.008 g g^{-1} to 3.065 μg g^{-1} for salmon, as shown in Table 4. LOQs ranged from 0.015 μg g^{-1} to 6.591 μg g^{-1} for tilapia and from 0.027 μg g^{-1} to 10.216 μg g^{-1} for salmon. LODs exhibited the same trend than observed for classical SPME,

513 and were higher for salmon. When SPME Arrow was used, the LODs for MP decreased by 23 and
514 13 times for tilapia and salmon, respectively. The results clearly show higher sensitivity can be
515 achieved with SPME Arrow when analyzing compounds with a wide range of polarities. In
516 addition, this work demonstrates that SPME Arrow can be used in direct-immersion mode to be
517 suitable for the analysis of organic compounds in complex matrices such as fish.

518 **3.5. Real sample analysis**

519 Eight fish samples (four tilapia and four salmon) obtained from local supermarkets were
520 analyzed to demonstrate robustness and suitability of the validated MSPD-SPME-GC-MS method.
521 The polar analytes (MP, PP, and DEET) were not detected in any of the samples, and this result is
522 logical because of the high LOD values obtained for these compounds. The bioaccumulation
523 potential of hydrophilic compounds with high water solubility is low in aquatic organisms (Streit,
524 1992) and they are frequently found in the ng g^{-1} range (Montesdeoca-Esponda et al., 2018).
525 Regarding the lipophilic compounds, only the musks (galaxolide and tonalide) were detected in all
526 samples, but the concentrations were below the LOQ. The results indicate that the significant
527 lipophilic (Log K_{ow} > 5, Table S1) and persistent nature of polycyclic musks aid in their tendency
528 to bioaccumulate in aquatic organisms like fish (Yao et al., 2018). This is an encouraging finding
529 due to the possible endocrine disruptive character of some of the PCPs studied, such as UV filters
530 and preservatives. In this particular study, the fact that most of the PCPs were not detected suggests
531 low human health risks associated with consumption of the analyzed fish.

532 The developed method was compared to other methods reported in the literature for PCPs
533 analysis in fish (Table S5). The proposed method is the only one dealing with five classes of PCPs
534 simultaneously. Most of the methods use from gram scale to hundreds of mg of sample, and the

proposed method uses only 25 mg due to the miniaturization. The precision was comparable with the other methods and the recoveries obtained were higher than most of the reported methods. The LODs were higher than most of the reported methods, but with the use of SPME Arrow, ng g-1 level was achieved, except for methyl paraben. Some alternatives to improve the sensitivity will be to perform on-fiber derivatization of the analytes with a silylating reagent, which is not environmentally friendly, or the use of more sensitive instrumentation, such as GC-MS/MS.

Conclusions

The method in this study was demonstrated to be accurate, sensitive, reliable, and easy to use for the determination of eleven PCPs of diverse groups in fish based on miniaturized MSPD combined with SPME followed by GC-MS. The use of Bondesil PPL as dispersant and a co-column with EMR-lipid sorbent to retain lipids makes the method suitable for the analysis of PCPs in various fish matrices, regardless of lipid content. The procedure has several notable advantages, in that it requires reduced sample and sorbent amounts, low volumes of organic solvent, and analyte no derivatization requirements. SPME Arrow permitted determination of all analytes at the ng g^{-1} level, demonstrating the method's high sensitivity without the need of MS/MS instrumentation. This method was applied in the determination of PCPs from commercially purchased fish. The overall approach is a suitable alternative for quality control and food safety purposes and in the screening of various PCPs in diverse fish species.

Acknowledgements

Iran Ocaña-Rios acknowledges the U.S.-Mexico Commission for Educational and Cultural Exchange (COMEXUS) for the Fulbright García-Robles Visiting Scholar Award. J.L.A.

556 acknowledges support from the Chemical Measurement and Imaging Program at the National
557 Science Foundation (CHE-2203891).

558 **CRediT authorship contribution statement**

559 **Iran Ocaña-Rios:** Conceptualization, Investigation, Methodology, Formal analysis,
560 Writing – original draft, Writing – review & editing. **Bhawana Thapa:** Validation, Investigation.
561 **Jared L. Anderson:** Conceptualization, Methodology, Writing – review & editing, Resources,
562 Visualization, Supervision, Project administration, Funding acquisition.

563 **Declaration of Competing Interest**

564 The authors declare that they have no known competing financial interests or personal
565 relationships that could have appeared to influence the work reported in this paper.

566 **References**

567 Agilent Technologies. (2017). *Agilent Captiva EMR—Lipid method guide*. Retrieved from
568 https://www.agilent.com/cs/library/usermanuals/public/5991-8308EN.pdf. Accessed January
569 7, 2023
570 Araujo, P. (2009). Key aspects of analytical method validation and linearity evaluation. Journal of
571 Chromatography B, 877, 2224-2234. https://doi.org/10.1016/j.jchromb.2008.09.030
572 Barker, S. A. (2000). Applications of matrix solid-phase dispersion in food analysis. Journal of
573 Chromatography A, 880, 63-68. https://doi.org/10.1016/S0021-9673(99)01290-X
574 Barker, S. A. (2007). Matrix solid phase dispersion (MSPD). *Journal of Biochemical and*
575 *Biophysical Methods*, 70, 151-162. https://doi.org/10.1016/j.jbbm.2006.06.005

576 Buchberger, W. W. (2011). Current approaches to trace analysis of pharmaceuticals and personal
577 care products in the environment. *Journal of Chromatography A*, 1218, 603-618.
578 https://doi.org/10.1016/j.chroma.2010.10.040
579 Campíns-Falcó, P., Verdú-Andrés, J., Sevillano-Cabeza, A., Molins-Legua, C., & Herráez-
580 Hernández, R. (2008). New micromethod combining miniaturized matrix solid-phase
581 dispersion and in-tube in-valve solid-phase microextraction for estimating polycyclic
582 aromatic hydrocarbons in bivalves. *Journal of Chromatography A*, 1211, 13-21.
583 https://doi.org/10.1016/j.chroma.2008.09.074
584 Canosa, P., Rodríguez, I., Rubí, E., Bollaín, M. H., & Cela, R. (2006). Optimisation of a solid-
585 phase microextraction method for the determination of parabens in water samples at the low
586 ng per litre level. *Journal of Chromatography A*, 1124, 3-10.
587 https://doi.org/10.1016/j.chroma.2006.03.045
588 Capriotti, A. L., Cavaliere, C., Giansanti, P., Gubbiotti, R., Samperi, R., & Laganà, A. (2010).
589 Recent developments in matrix solid-phase dispersion extraction. *Journal of*
590 *Chromatography A,* 1217, 252-2532. https://doi.org/10.1016/j.chroma.2010.01.030
591 Castro, O., Trabalón, L., Schilling, B., Borrull, F., & Pocurull, E. (2019). Solid phase
592 microextraction Arrow for the determination of synthetic musk fragrances in fish samples.
593 *Journal of Chromatography A*, 1591, 55-61. https://doi.org/10.1016/j.chroma.2019.01.032
594 Chen, F., Gong, Z., & Kelly, B. C. (2017). Bioaccumulation behavior of pharmaceuticals and
595 personal care products in adult zebrafish (*Danio rerio*): Influence of physical-chemical
596 properties and biotransformation. *Environmental Science & Technology*, 51, 11085-11095.
597 https://doi.org/10.1021/acs.est.7b02918

598 Eguiraun, H., Izagirre, U., & Martinez, I. (2015). A paradigm shift in safe seafood production:
599 From contaminant detection to fish monitoring - Application of biological warning systems
600 to aquaculture. *Trends in Food Science & Technology*, 43, 104-113.
601 https://doi.org/10.1016/j.tifs.2015.01.007
602 Gionfriddo, E., Gruszecka, D., Li, X., & Pawliszyn, J. (2020). Direct-immersion SPME in soy
603 milk for pesticide analysis at trace levels by means of a matrix-compatible coating. *Talanta*,
604 211, Article 120746. https://doi.org/10.1016/j.talanta.2020.120746
605 Herrington, J. S., Gómez-Ríos, G. A., Myers, C., Stidsen, G., & Bell, D. S. (2020). Hunting
606 molecules in complex matrices with SPME Arrows: A review. *Separations*, 7, Article 12.
607 https://doi.org/10.3390/separations7010012
608 Hopkins, Z. R., & Blaney, L. (2016). An aggregate analysis of personal care products in the
609 environment: Identifying the distribution of environmentally-relevant concentrations.
610 *Environment International*, 92-93, 301-316. http://dx.doi.org/10.1016/j.envint.2016.04.026
611 Jakimska, A., Huerta, B., Barganska, Z., Kot-Wasik, A., Rodríguez-Mozaz, S., & Barceló, D.
612 (2013). Development of a liquid chromatography-tandem mass spectrometry procedure for
613 determination of endocrine disrupting compounds in fish from Mediterranean rivers. *Journal*
614 *of Chromatography A*, 1306, 44-58. https://doi.org/10.1016/j.chroma.2013.07.050
615 Jesus, A., Augusto, I., Duarte, J., Sousa, E., Cidade, H., Cruz, M. T., Sousa Lobo, J. M., &
616 Almeida, I. F. (2022). Recent Trends on UV filters. *Applied Sciences*, 12, Article 12003.
617 https://doi.org/10.3390/app122312003
618 Kataoka, H., Lord, H. L., & Pawliszyn, J. (2000). Applications of solid-phase microextraction in
619 food analysis. *Journal of Chromatography A*, 880, 35-62. https://doi.org/10.1016/S0021-
620 9673(00)00309-5

621 Kim, J. W., Ramaswamy, B. R., Chang, K. H., Isobe, T., & Tanabe, S. (2011). Multiresidue
622 analytical method for the determination of antimicrobials, preservatives, benzotriazole UV
623 stabilizers, flame retardants and plasticizers in fish using ultra high performance liquid
624 chromatography coupled with tandem mass spectrometry. *Journal of Chromatography A*,
625 1218 3511-3520. https://doi.org/10.1016/j.chroma.2011.04.006
626 Kristenson, E. M., Ramos, L., & Brinkman, U. A. T. (2006). Recent advances in matrix solid-
627 phase dispersion. *Trends in Analytical Chemistry*, 25, 96-111.
628 https://doi.org/10.1016/j.trac.2005.05.011
629 Li, Y., Harir, M., Lucio, M., Kanawati, B., Smirnov, K., Flerus, R., Koch, B. P., Schmitt-
630 Kopplin, P., & Hertkorn, N. (2016). Proposed guidelines for solid phase extraction of
631 Suwannee river dissolved organic matter. *Analytical Chemistry*, 88, 6680-6688.
632 https://doi.org/10.1021/acs.analchem.5b04501
633 Meinerling, M., & Daniels, M. (2006). A validated method for the determination of traces of UV
634 filters in fish using LC–MS/MS. *Analytical and Bioanalytical Chemistry*, 386, 1465-1473.
635 https://doi.org/10.1007/s00216-006-0706-9
636 Moliner-Martinez, Y., Campíns-Falcó, P., Molins-Legua, C., Segovia-Martínez, L., & Seco-
637 Torrecillas, A. (2009). Miniaturized matrix solid phase dispersion procedure and solid phase
638 microextraction for the analysis of organochlorinated pesticides and polybrominated
639 diphenylethers in biota samples by gas chromatography electron capture detection. *Journal*
640 *of Chromatography A*, 1216, 6741-6745. https://doi.org/10.1016/j.chroma.2009.08.021
641 Montesdeoca-Esponda, S., Checchini, L., Bubba, M. D., Sosa-Ferrera, Z., & Santana-Rodriguez,
642 J. J. (2018). Analytical approaches for the determination of personal care products and

643 evaluation of their occurrence in marine organisms. *Science of the Total Environment*, 633,
644 405-425. https://doi.org/10.1016/j.scitotenv.2018.03.182
645 Moradi, Y., Bakar, J., Motalebi, A. A., Syed Muhamad S. H., & Che Man, Y. (2011). A review
646 on fish lipid: Composition and changes during cooking methods. *Journal of Aquatic Food*
647 *Product Technology*, 20, 379-390, https://doi.org/10.1080/10498850.2011.576449
648 Mottaleb, M. A., Usenko, S., O'Donnell, J. G., Ramirez, A. J., Brooks, B. W., & Chambliss, C.
649 K. (2009). Gas chromatography–mass spectrometry screening methods for select UV filters,
650 synthetic musks, alkylphenols, an antimicrobial agent, and an insect repellent in fish.
651 *Journal of Chromatography A*, 1216 815-823. https://doi.org/10.1016/j.chroma.2008.11.072
652 Negreira, N., Rodríguez, I., Rodil, R., Rubí, E., & Cela, R. (2013). Optimization of matrix solid-
653 phase dispersion conditions for UV filters determination in biota samples. *International*
654 *Journal of Environmental Analytical Chemistry*, 93, 1174-1188,
655 https://doi.org/10.1080/03067319.2012.702277
656 Núñez, M., Borrull, F., Pocurull, E., & Fontanals, N. (2017). Sample treatment for the
657 determination of emerging organic contaminants in aquatic organisms. *Trends in Analytical*
658 *Chemistry*, 97 136-145. http://dx.doi.org/10.1016/j.trac.2017.09.007
659 Pawliszyn, J. (2012). Theory of Solid-Phase Microextraction. In J. Pawliszyn (Ed.), *Handbook of*
660 *Solid Phase Microextraction* (pp. 13-59), Elsevier. https://doi.org/10.1016/B978-0-12-
661 416017-0.00002-4
662 Peng, X., Jin, J., Wang, C., Ou, W., & Tang, C. (2015). Multi-target determination of organic
663 ultraviolet absorbents in organism tissues by ultrasonic assisted extraction and ultra-high
664 performance liquid chromatography tandem mass spectrometry. *Journal Chromatography A*,
665 1384, 97-106. https://doi.org/10.1016/j.chroma.2015.01.051

Portillo-Castillo, O. J., Castro-Ríos, R., Chávez-Montes, A., González-Horta, A., Cavazos-Rocha, N., Waksman de Torres, N. H., & Garza-Tapia, M. (2018). Developments of solid-phase microextraction fiber coatings for environmental pharmaceutical and personal care products analysis. *Reviews in Analytical Chemistry*, 37, Article 20170018. https://doi.org/10.1515/revac-2017-0018

Ramirez, A. J., Brain, R. A., Usenko, A. J. S., Mottaleb, M. A., O'Donnell, J. G., Stahl, L. L., Wathen, J. B., Snyder, B. D., Pitt, J. L., Perez-Hurtado, P., Dobbins, L. L., Brooks, B. W., & Chambliss, C. K. (2009). Occurrence of pharmaceuticals and personal care products in fish: results of a national pilot study in the United States. *Environmental Toxicology and Chemistry*, 28, 2587-2597. https://doi.org/10.1897/08-561.1

Sauvé, S., & Desrosiers, M. (2014). A review of what is an emerging contaminant. *Chemistry Central Journal*, 8, Article 15. https://doi.org/10.1186/1752-153X-8-15

Streit, B. (1992). Bioaccumulation processes in ecosystems. *Experientia*, 48, 955-970. https://doi.org/10.1007/BF01919142

Tsai, D. Y., Chen, C. L., & Ding, W. H. (2014). Optimization of matrix solid-phase dispersion for the rapid determination of salicylate and benzophenone-type UV absorbing substances in marketed fish. *Food Chemistry*, 154, 211-216. http://dx.doi.org/10.1016/j.foodchem.2014.01.013

Urban, M., & Lesueur, C. (2017). Comparing d-SPE sorbents of the QuEChERS extraction method and EMR-Lipid for the determination of polycyclic aromatic hydrocarbons (PAH4) in food of animal and plant origin. *Food Analytical Methods*, 10, 2111-2124. https://doi.org/10.1007/s12161-016-0750-9

688 Wianowska, D., Gil, M., & Olszowy, M. (2020). Miniaturized methods of sample preparation. In
689 C. M. Hussain (Ed.), Handbook on Miniaturization in Analytical Chemistry (pp. 99-125).
690 Elsevier Inc. https://doi.org/10.1016/B978-0-12-819763-9.00005-2
691 Witorsch, R. J., & Thomas, J. A. (2010). Personal care products and endocrine disruption: A
692 critical review of the literature. *Critical Reviews in Toxicology*, 40, 1-30.
693 https://doi.org/10.3109/10408444.2010.515563
694 Xiao, S., Lv, S., Cheng, Z., Hu, C., Li, P., Nan, F., Liu, X., Liu, D., Zhou, Z., & Wang, P.
695 (2021). A simple method for the determination of pharmaceutical and personal care products
696 in fish tissue based on matrix solid-phase dispersion. *Journal of Agricultural and Food*
697 *Chemistry*, 69, 15738-15745. https://doi.org/10.1021/acs.jafc.1c05232
698 Yao, L., Zhao, J. L., Liu, Y. S., Zhang, Q. Q., Jiang, Y. X., Liu, S., Liu, W. R., Yang, Y. Y., &
699 Ying, G. G. (2018). Personal care products in wild fish in two main Chinese rivers:
700 Bioaccumulation potential and human health risks. *Science of The Total Environment*, 621,
701 1093-1102. https://doi.org/10.1016/j.scitotenv.2017.10.117
702 Yin, Z., Yuan, B., Lyu, W., Huang, Q., Simon, J. E., & Wu, Q. (2022). Method development and
703 validation for analysis of phenolic compounds in fatty complex matrices using enhanced
704 matrix removal (EMR) lipid cleanup and UHPLC-QqQ-MS/MS. *Food Chemistry*, 373,
705 Article 131096. https://doi.org/10.1016/j.foodchem.2021.131096
706

Table 1. Validation results of the MSPD-SPME-GC-MS method for tilapia samples (n=3)

Analyte	Linearity[a]					Accuracy (%Recovery)				Precision (repeatability, RSD %)			
						Concentration (μg g^{-1}, dry weight)[c]				Concentration (μg g^{-1}, dry weight)[c]			
	Slope	SE[b]	Y-intercept	SE[b]	R^2	5	10	100	300	5	10	100	300
MP	$6.77 \cdot 10^{-6}$	$5.78 \cdot 10^{-7}$	$3.57 \cdot 10^{-4}$	$1.92 \cdot 10^{-4}$	0.984	-	-	108.63	99.91	-	-	5.20	8.71
DEET	$1.12 \cdot 10^{-4}$	$3.36 \cdot 10^{-6}$	$1.81 \cdot 10^{-3}$	$9.45 \cdot 10^{-4}$	0.993	-	98.36	85.79	86.54	-	9.31	10.02	10.92
PP	$1.84 \cdot 10^{-4}$	$3.90 \cdot 10^{-6}$	$-5.61 \cdot 10^{-4}$	$1.10 \cdot 10^{-3}$	0.999	-	99.74	88.09	93.90	-	4.67	5.77	9.65
GAL	$1.49 \cdot 10^{-3}$	$4.50 \cdot 10^{-5}$	$4.07 \cdot 10^{-2}$	$1.27 \cdot 10^{-2}$	0.988	98.22	98.94	87.08	80.44	9.81	10.20	5.76	3.25
TON	$1.41 \cdot 10^{-3}$	$3.96 \cdot 10^{-5}$	$3.92 \cdot 10^{-2}$	$1.11 \cdot 10^{-2}$	0.987	95.86	98.22	89.61	80.31	11.82	4.36	5.89	2.69
CP	$3.27 \cdot 10^{-3}$	$7.83 \cdot 10^{-5}$	$2.48 \cdot 10^{-2}$	$2.20 \cdot 10^{-2}$	0.996	96.54	96.91	88.95	83.39	8.02	2.49	2.83	7.85
OXY	$4.90 \cdot 10^{-3}$	$2.00 \cdot 10^{-4}$	$7.44 \cdot 10^{-2}$	$5.63 \cdot 10^{-2}$	0.991	92.47	92.59	91.17	85.24	10.06	7.14	3.17	5.44
4-MBC	$1.53 \cdot 10^{-3}$	$5.29 \cdot 10^{-5}$	$4.03 \cdot 10^{-2}$	$1.49 \cdot 10^{-2}$	0.985	93.69	94.81	92.22	81.26	9.45	3.21	3.30	4.96
TCS	$1.71 \cdot 10^{-3}$	$7.44 \cdot 10^{-5}$	$3.61 \cdot 10^{-2}$	$2.09 \cdot 10^{-2}$	0.980	97.43	92.19	92.70	84.49	7.38	5.22	6.42	5.39
EHPABA	$1.08 \cdot 10^{-2}$	$3.77 \cdot 10^{-4}$	$1.82 \cdot 10^{-1}$	$1.06 \cdot 10^{-1}$	0.987	94.29	93.87	85.98	80.11	8.55	6.67	8.60	2.59
OCT	$4.38 \cdot 10^{-4}$	$2.03 \cdot 10^{-5}$	$8.39 \cdot 10^{-3}$	$5.70 \cdot 10^{-3}$	0.972	-	98.55	87.59	89.17	-	10.56	12.04	6.36

[a] Range of linearity: 150-500 μg g^{-1} for MP (n=15) and 10-500 μg g^{-1} for the rest of the analytes (n=21)

[b] Standard error of the slope and the Y-intercept

[c]The values not reported were < LOQ of the analyte

Table 2. Validation results of the MSPD-SPME-GC-MS method for salmon samples (n=3)

Analyte	Linearity[a]					Accuracy (%Recovery)				Precision (repeatability, RSD %)			
						Concentration (μg g^{-1}, dry weight)[c]				Concentration (μg g^{-1}, dry weight)[c]			
	Slope	SE[b]	Y-intercept	SE[b]	R^2	5	10	100	300	5	10	100	300
MP	$9.91 \cdot 10^{-6}$	$8.09 \cdot 10^{-7}$	$6.24 \cdot 10^{-4}$	$2.68 \cdot 10^{-4}$	0.971	-	-	105.98	107.39	-	-	11.94	5.54
DEET	$1.42 \cdot 10^{-4}$	$6.84 \cdot 10^{-6}$	$2.41 \cdot 10^{-3}$	$1.92 \cdot 10^{-3}$	0.987	-	99.83	88.55	89.36	-	9.06	12.01	1.82
PP	$2.06 \cdot 10^{-4}$	$1.74 \cdot 10^{-5}$	$2.97 \cdot 10^{-3}$	$4.90 \cdot 10^{-3}$	0.982	-	101.13	89.65	82.41	-	6.16	9.08	1.62
GAL	$1.48 \cdot 10^{-3}$	$6.03 \cdot 10^{-5}$	$-6.38 \cdot 10^{-4}$	$1.69 \cdot 10^{-2}$	0.986	98.91	98.11	87.03	90.94	10.23	10.11	12.96	5.29
TON	$1.44 \cdot 10^{-3}$	$5.76 \cdot 10^{-5}$	$-2.33 \cdot 10^{-3}$	$1.62 \cdot 10^{-2}$	0.985	97.32	96.60	85.67	89.68	9.82	9.19	12.01	5.21
CP	$4.30 \cdot 10^{-3}$	$8.89 \cdot 10^{-5}$	$-8.96 \cdot 10^{-3}$	$2.50 \cdot 10^{-2}$	0.998	98.36	93.94	83.13	83.95	7.25	4.86	3.30	8.22
OXY	$6.44 \cdot 10^{-3}$	$3.15 \cdot 10^{-4}$	$9.54 \cdot 10^{-2}$	$8.85 \cdot 10^{-2}$	0.975	93.27	100.79	89.11	85.23	10.56	9.68	7.73	8.05
4-MBC	$1.93 \cdot 10^{-3}$	$6.07 \cdot 10^{-5}$	$4.52 \cdot 10^{-3}$	$1.71 \cdot 10^{-2}$	0.993	95.32	96.72	85.51	90.66	9.59	10.17	8.27	5.82
TCS	$2.36 \cdot 10^{-3}$	$8.58 \cdot 10^{-5}$	$9.73 \cdot 10^{-3}$	$2.41 \cdot 10^{-2}$	0.991	95.52	106.41	94.19	97.32	8.71	9.51	9.29	4.87
EHPABA	$1.48 \cdot 10^{-2}$	$6.90 \cdot 10^{-4}$	$5.30 \cdot 10^{-3}$	$1.94 \cdot 10^{-1}$	0.985	95.31	99.93	93.11	84.13	9.32	8.34	7.98	4.22
OCT	$9.34 \cdot 10^{-4}$	$4.61 \cdot 10^{-5}$	$8.90 \cdot 10^{-4}$	$1.30 \cdot 10^{-2}$	0.973	-	101.77	94.83	82.19	-	6.73	6.27	4.48

[a] Range of linearity: 150-500 μg g^{-1} for MP (n=15) and 10-500 μg g^{-1} for the rest of the analytes (n=21)

[b] Standard error of the slope and the Y-intercept

[c]The values not reported were < LOQ of the analyte

Table 3. LODs and LOQs for the MSPD-SPME-GC-MS method for tilapia and salmon samples

Analyte	Dry weight (μg g^{-1})				Wet weight (μg g^{-1})			
	Tilapia		Salmon		Tilapia		Salmon	
	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]
MP	44.95	149.83	39.23	130.77	10.46	34.87	14.36	47.87
DEET	2.26	7.53	2.76	9.21	0.53	1.75	1.01	3.37
PP	1.68	5.60	1.81	6.05	0.39	1.30	0.66	2.21
GAL	0.10	0.34	0.21	0.70	0.02	0.08	0.08	0.26
TON	0.09	0.29	0.18	0.59	0.02	0.07	0.07	0.22
CP	0.05	0.18	0.06	0.21	0.01	0.04	0.02	0.08
OXY	0.04	0.12	0.04	0.13	0.01	0.03	0.01	0.05
4-MBC	0.09	0.28	0.12	0.41	0.02	0.07	0.04	0.15
TCS	0.09	0.31	0.11	0.36	0.02	0.07	0.04	0.13
EHPABA	0.06	0.21	0.08	0.28	0.01	0.05	0.03	0.10
OCT	2.03	6.77	1.76	5.87	0.47	1.58	0.64	2.15

[a] Limit of detection, calculated as the concentration corresponding to 3 times the signal-to-noise ratio

[b] Limit of quantitation, calculated as the concentration corresponding to 10 times the signal-to-noise ratio

Table 4. LODs and LOQs for the MSPD-SPME Arrow-GC-MS method for tilapia and salmon samples

Analyte	Dry weight (μg g^{-1})				Wet weight (μg g^{-1})			
	Tilapia		Salmon		Tilapia		Salmon	
	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]	LOD[a]	LOQ[b]
MP	1.977	6.591	3.065	10.216	0.460	1.534	1.122	3.740
DEET	0.061	0.204	0.139	0.462	0.014	0.048	0.051	0.169
PP	0.086	0.285	0.170	0.565	0.020	0.066	0.062	0.207
GAL	0.005	0.015	0.008	0.027	0.001	0.004	0.003	0.010
TON	0.005	0.015	0.009	0.029	0.001	0.004	0.003	0.011
CP	0.004	0.015	0.008	0.028	0.001	0.003	0.003	0.010
OXY	0.005	0.017	0.008	0.027	0.001	0.004	0.003	0.010
4-MBC	0.010	0.032	0.021	0.071	0.002	0.007	0.008	0.026
TCS	0.009	0.031	0.015	0.050	0.002	0.007	0.006	0.018
EHPABA	0.010	0.033	0.012	0.039	0.002	0.008	0.004	0.014
OCT	0.149	0.498	0.079	0.265	0.035	0.116	0.029	0.097

[a] Limit of detection, calculated as the concentration corresponding to 3 times the signal-to-noise ratio

[b] Limit of quantitation, calculated as the concentration corresponding to 10 times the signal-to-noise ratio

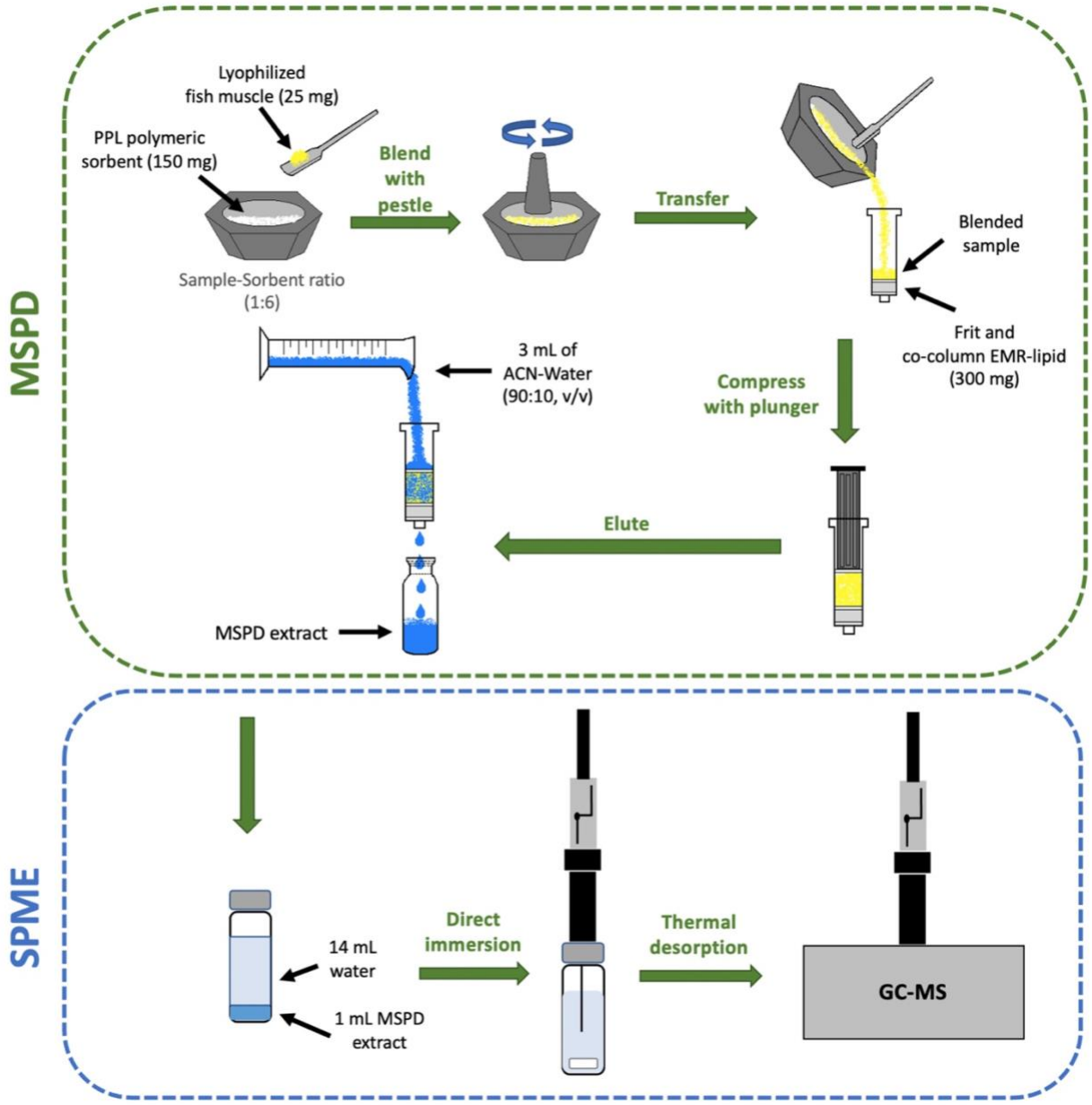

Figure 1. Optimized procedure for the MSPD-SPME-GC-MS method

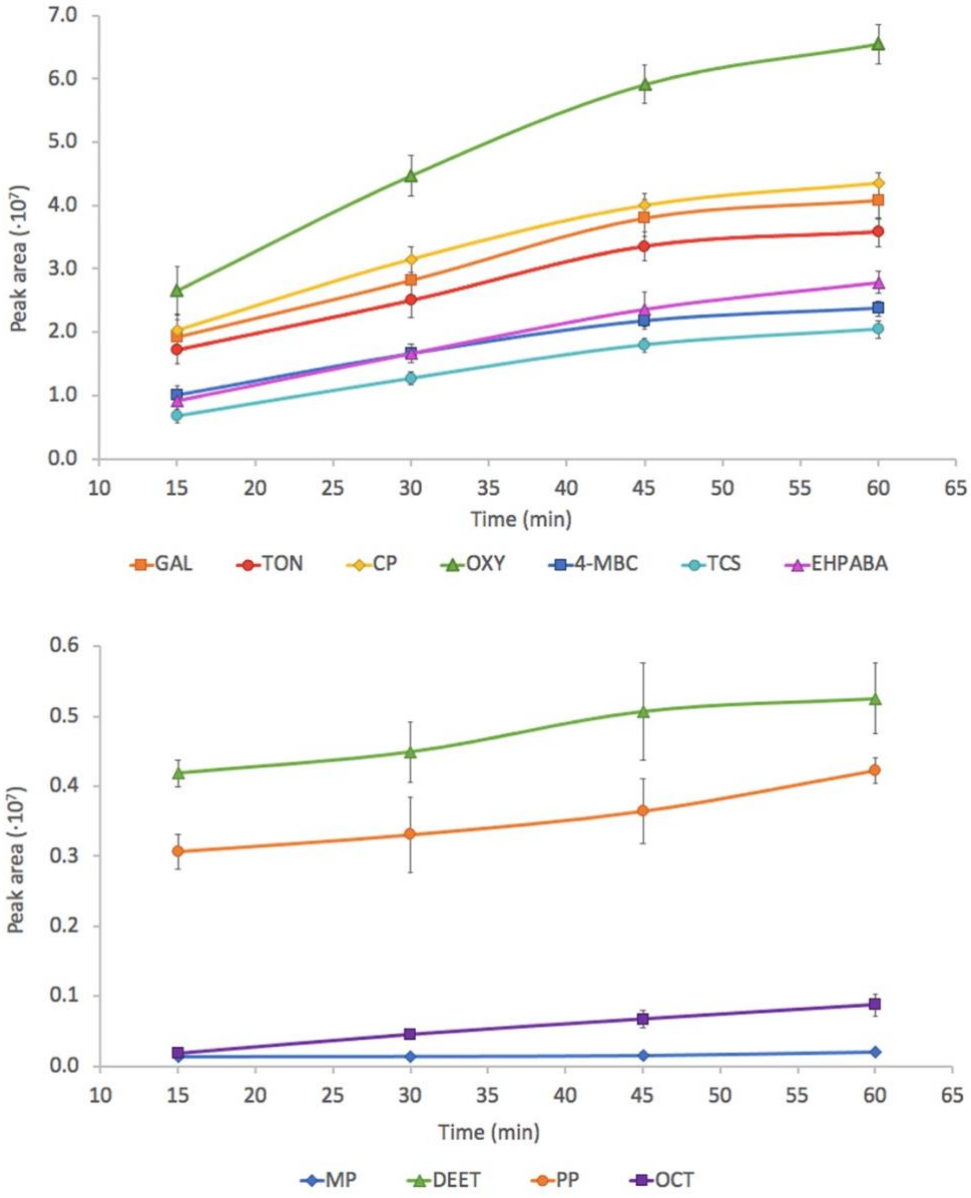

Figure 2. Extraction time profiles obtained for the SPME-GC-MS method using a PDMS/DVB fiber (n=3). Experimental conditions: 15 mL of water containing the analytes at 100 ng mL^{-1}, ACN 5 % (v/v), NaCl 10 % (w/v), extraction temperature 35 °C, stirring rate 1000 rpm.