Methodology for Analysis of UV Filters in Tilapia Using Off-line MSPD Followed by On-line SPE–LC/UV

José J. Olmos-Espejel, Óscar E. Mogica-García, Gerson J. Duran-Gasca & Magaly L. Carmona-López

Chromatographia
An International Journal for Separation Science

ISSN 0009-5893

Chromatographia
DOI 10.1007/s10337-020-03867-2

Springer

Your article is protected by copyright and all rights are held exclusively by Springer-Verlag GmbH Germany, part of Springer Nature. This e-offprint is for personal use only and shall not be self-archived in electronic repositories. If you wish to self-archive your article, please use the accepted manuscript version for posting on your own website. You may further deposit the accepted manuscript version in any repository, provided it is only made publicly available 12 months after official publication or later and provided acknowledgement is given to the original source of publication and a link is inserted to the published article on Springer's website. The link must be accompanied by the following text: "The final publication is available at link.springer.com”.

Springer

Author's personal copy

Chromatographia
https://doi.org/10.1007/s10337-020-03867-2

ORIGINAL

Methodology for Analysis of UV Filters in Tilapia Using Off-line MSPD Followed by On-line SPE–LC/UV

José J. Olmos-Espejel[1] · Óscar E. Mogica-García[1] · Gerson J. Duran-Gasca[1] · Magaly L. Carmona-López[1]

Received: 20 November 2019 / Revised: 26 January 2020 / Accepted: 27 January 2020
© Springer-Verlag GmbH Germany, part of Springer Nature 2020

Abstract

A sensitive method was developed and validated for the determination of UV filters (oxybenzone, octocrylene, 4-methylbenzylidene camphor, 2-ethylhexyl-4-methoxycinnamate and avobenzone) in tilapia muscle using matrix solid-phase dispersion followed by on-line solid-phase extraction–liquid chromatography/UV. The validation showed good linearity from 100 to 3600 ng g^{-1} with r^2 values > 0.98. Precision values with relative standard deviations were < 24% for all analytes. The accuracy was evaluated with the recovery of spiked muscle samples, and it ranged between 89 and 96% for most analytes, except 2-ethylhexyl-4-methoxycinnamate. A positive matrix effect was observed for oxybenzone, so a matrix-matched calibration curve was used for quantitation purposes. This methodology was applied to incurred samples of fish exposed to the analytes for 3 days (100 µg L^{-1} each). Residual quantities of the contaminants were measured after 24, 48 and 72 h of exposure, and concentrations ranged between 400 and 2900 ng g^{-1}. The developed method was able to quantify all of the target compounds at trace levels.

Keywords Liquid chromatography · On-line solid-phase extraction · Matrix solid-phase dispersion · Fish muscle · UV filters

Introduction

In the last two decades, UV filters (UVFs) have been extensively used in a variety of personal care products, such as sunscreen, moisturizing lotion, and lipstick, among others, to mitigate the harmful effects of ultraviolet (UV) radiation in skin cancer development, as well as for erythema and photoaging. Additionally, they are used in many industrial goods, such as plastics, textiles, and food packaging, to protect polymers and pigments against photoinduced degradation. UVFs enter the aquatic environment in two different ways: by direct inputs of recreational activities, such as sloughing off human bodies during swimming, or through indirect inputs, such as the discharge of wastewater treatment plants [1–3].

There are approximately 42 different organic compounds that can be employed as UV filters in sunscreen products according to legislation in place in the European Union, USA and Japan [4]. Oxybenzone, octocrylene, 4-methylbenzylidene camphor, 2-ethylhexyl-4-methoxycinnamate and avobenzone are the most frequently used UV filters in sunscreen formulations [3], and all of them present effects as endocrine disruptors or promotors (in vitro) of growth for certain breast cancer cells [2].

The enormous production and use of UVFs have caused an increase in their concentration in the aquatic environment and a greater potential risk of bioaccumulation and biomagnification in several species of freshwater and saltwater fish [5]. Several ecological factors are involved in the degree of UVF absorption by aquatic species, including size (weight and length), body lipid content and sampling location [1].

Aquatic organisms are considered good biomonitors because they absorb foreign chemicals. In fish, the contaminant concentrations depend on the amounts of analyte in contact with the organism, the rate of uptake (through diet and respiration) and elimination (by metabolism, egestion, respiration and growth) [6]. Among these, tilapia (*Oreochromis urolepis hornorum*) has been used in toxicological

✉ José J. Olmos-Espejel
j-olmos@comunidad.unam.mx

[1] Facultad de Estudios Superiores Cuautitlán, Sección de Química Analítica, Universidad Nacional Autónoma de México, Av. Primero de Mayo S/N, Santa María de Guadalupe Las Torres, Campo Uno, C.P. 54740, Cuautitlán Izcalli, Estado de México, Mexico

Published online: 08 February 2020

Author's personal copy

studies to evaluate the effects of contaminants, such as fungicides, organophosphorus pesticides and heavy metals. Moreover, these organisms are resistant to different stress conditions [7].

UVFs have been detected at concentrations between 9 and 2400 ng g^{-1} [8, 9] in fish samples. Despite their low concentration, these levels have the potential for transcriptional alterations and endocrine disruption in species such as rats, fish and frogs [10].

For this reason, several methods have been developed for the determination of UVFs in fish tissues at trace levels, many of which used techniques such as Soxhlet, solid–liquid and pressurized liquid extraction, with an additional clean-up step (gel permeation chromatography, silica or florisil columns) [1]. Appropriate sample preparation procedure is required because the samples often contain low amounts of UVFs and a large quantity of interfering compounds that make analysis difficult [11, 12].

Among sample preparation techniques, matrix solid-phase dispersion (MSPD) is suitable for solid and semisolid samples due to the direct mechanical blending of the sample with an abrasive and appropriate solid support material, mainly florisil and C18-bonded silica, which is then packed into a polypropylene syringe that contains a clean-up sorbent to retain coextracted interfering species [13, 14]. The success of this technique lies in its simplicity, affordability, flexibility and robustness. Furthermore, the implemented conditions help to prevent the degradation of analytes, and they provide both extraction and clean-up in a single step [15].

In a typical analysis of contaminants in biota, after the sample preparation step, the analysis is performed with a chromatographic system (LC or GC) to isolate target analytes from any remaining interferences. This can be done with either off-line systems or physically connected (on-line) systems. In off-line mode, the volume of extract is commonly reduced to improve the sensitivity of the method because a few microliters of the extract is introduced into the chromatographic system. In on-line mode, there is a complete coupling between the sample preparation step and chromatographic system, allowing for the preconcentration of the analytes and its subsequent introduction into the system in a process equivalent to injecting the entire volume of the extract. On-line systems are suitable for automation, and they provide an improvement in precision and detection limit at low concentration levels [16]. Solid-phase extraction (SPE) is a technique commonly used in on-line mode; it involves the extraction of analytes by a sorbent phase, and it has made the development of faster and more sensitive methods possible by reducing analysis time and sample contamination [17, 18]. On-line SPE has been successfully used for the determination of contaminants in liquid matrices, such as pharmaceuticals in blood [19], insect repellent and its metabolites in urine [20], ochratoxin A in wine [21], tetracyclines in honey [22] and benzotriazoles in water [23]. UVFs have also been successfully analyzed using on-line SPE coupled to liquid chromatography in liquid samples, such as natural waters and wastewaters [24], swimming pool water and seawater [25], surface water [26] and urine [27, 28].

There is scarce information about the use of on-line SPE for solid sample analysis because of the matrix complexity. These methods require an additional sample preparation technique to obtain a clean extract prior to on-line analysis. For this purpose, matrix solid-phase dispersion (MSPD) and solid–liquid extraction have been used, particularly MSPD coupled with on-line SPE–LC, to analyze polycyclic aromatic hydrocarbons in animal tissue [29, 30].

The aim of this work was to develop an off-line MSPD, followed by an on-line SPE–LC/UV method for the determination of five UVFs (oxybenzone, octocrylene, 4-methylbenzylidene camphor, 2-ethylhexyl-4-methoxycinnamate and avobenzone) in tilapia muscle.

Materials and Methods

The properties of the UVFs are listed in Electronic Supplementary Material Table S1. The analytical standards of oxybenzone (OXY), octocrylene (OCT), 4-methylbenzylidene camphor (MBC), 2-ethylhexyl-4-methoxycinnamate (EHMC) and avobenzone (AVO) were purchased from Sigma-Aldrich (purity > 98%). Individual stock solutions at 2000 µg mL^{-1} were prepared in acetonitrile. A standard solution at 500 µg mL^{-1} containing the five UVFs was prepared with acetonitrile and stored in the dark at 4 °C. Water (18.2 MΩ cm resistivity) was obtained from a Millipore Simplicity UV water purification system (Bedford, MA, USA). For MSPD, C18 silica (particle diameter 40–60 µm), 6 mL polypropylene SPE cartridges and polyethylene frits were obtained from Agilent Technologies (Santa Clara, CA, USA). High-performance ZORBAX guard fittings kit and a ZORBAX Eclipse Plus guard cartridge (95 Å C18, 5 µm, 4.6 × 12.5 mm) were acquired from Agilent Technologies. HPLC-grade methanol and acetonitrile (ACN) were purchased from J.T. Baker (Phillipsburg, NJ, USA). To avoid cross-contamination, all glassware was cleaned overnight with an aqueous 5% HNO_3 solution (v/v) and was then rinsed with distilled water and 1 mL of ACN.

Fish Samples

To optimize the MSPD methodology, tilapia fillets (*Oreochromis urolepis hornorum*) were acquired from a local market. Fish muscle was homogenized in a food processor and lyophilized.

Author's personal copy

Additionally, juvenile tilapia obtained from the Faculty of Veterinary Medicine and Zootechnics, UNAM, were used as incurred samples to test the method. They were acclimated to laboratory conditions in an aerated aquarium for a week (25–27 °C, pH = 7, 10/14 h light/dark photoperiod, dissolved oxygen: 4 mg L^{-1}), and water was renewed every 72 h.

Off-line MSPD Methodology

First, 0.5 g of lyophilized muscle and 1 g of C18 silica (preconditioned with 5 mL of methanol and vacuum dried) were blended in an agate mortar until a homogeneous mixture was obtained. The mixture was placed into a 6 mL polypropylene cartridge with a polyethylene frit in the bottom, compressed and covered with another polyethylene frit. Analytes were eluted from the cartridge with 5 mL of acetonitrile. The extract was centrifuged at 4000 rpm for 15 min, and the entire supernatant was diluted with 10 mL of deionized water for subsequent preconcentration and analysis on the on-line SPE–LC/UV system. A summary of the methodology is shown in Electronic Supplementary Material Fig. S1.

Preparation of Matrix-Matched Standards

Blank MSPD extracts (5 mL) obtained from unspiked fish muscle were used to prepare matrix-matched standards. Different amounts of each analyte were added to the final ACN extracts at six concentration levels (10, 40, 80, 160, 250 and 360 ng mL^{-1}). To construct a calibration curve, the matrix-matched standards were diluted with 10 mL of deionized water and analyzed with the on-line SPE–LC/UV system. This calibration curve was used for quantitation purposes.

On-line SPE–LC/UV Analysis

The UVFs were analyzed on a 1100 liquid chromatograph with a quaternary pump (model G1311A) coupled to a UV diode array detector (DAD, model G1315B) from Agilent Technologies (Santa Clara, CA, USA). The LC system was equipped with an autosampler (model G1313A) and temperature control module (model G1316A). The direction of the mobile phase flow was controlled by an automatic rheodyne valve (Berkeley, CA, USA) connected to the LC temperature control module. Samples were preconcentrated on a ZORBAX Eclipse Plus C18 guard cartridge using an Eldex Optos Series 2SM pump (Napa, CA, USA). An inline filter (4.8 mm, 2 µm) was connected between the Eldex pump and precolumn to extend the useful life of the preconcentration column. The diluted extracts were loaded using the preconcentration pump (PP) at a constant flow of 1.8 mL min^{-1}. Table 1 shows the operating conditions used for the on-line system.

Analytical separation was performed on an SB-C18 analytical column (250 mm × 4.6 mm I.D., 5 µm particle size) supplied by Agilent Technologies in isocratic elution mode (85% methanol and 15% water) at a flow rate of 1 mL min^{-1}. The detection wavelengths selected for each compound were as follows: OXY = 290 nm; OCT, MBC and EHMC = 310 nm; and AVO = 360 nm. A total run time of 20 min allowed for the separation of the five UVFs from interfering compounds. Data analysis was performed using ChemStation software version 10.02.

Method Validation

The following parameters were evaluated: precision (repeatability), accuracy, limits of detection (LODs), limits of quantitation (LOQs) and linearity. Precision was evaluated in terms of repeatability by analyzing six extracts of spiked samples at 400 and 1200 ng g^{-1} on two different days. For each concentration, triplicate analysis was conducted, and the relative standard deviation (RSD) was calculated. The linearity was evaluated by analyzing muscle samples spiked at six concentration levels (100, 400, 800, 1600, 2500 and 3600 ng g^{-1}). Three sample replicates were made at each concentration level. The determination coefficients (r^2), slopes and Y intercepts of the calibration curve were calculated for each analyte. The accuracy was evaluated with the slope of the curve obtained by plotting the recovered amount of each analyte versus the amount initially added to the spiked samples. The experimental LODs and LOQs

Table 1 Conditions for on-line SPE–LC/UV system

Activity	Pump used	Switching valve position	Solvent applied	Flow mL min^{-1}
Precolumn conditioning	PP	Load	a) 5 mL acetonitrile 100% b) 5 mL water 100%	1.8
Sample loading	PP	Load	15 mL extract diluted	1.8
On-line cleanup	PP	Load	2 mL water 100%	1.8
analyte separation	CP	Injection[a]	Mobile phase MeOH:water (85:15, v/v)	1.0

PP preconcentration pump, *CP* chromatographic pump

[a]Maintained during 6 min of analysis before reconditioning of the precolumn

Author's personal copy

were determined with signal-to-noise ratios (S/N) of 3 and 10, respectively.

Incurred Fish Samples

Ten juvenile tilapia (length 18.5 ± 3.0 cm; weight 39.8 ± 5.6 g) were used as incurred samples. One fish was placed in an aerated aquarium with 30 L of potable water as a control. Nine fish were placed in another aerated aquarium with 70 L of spiked water at 100 ng mL^{-1} of each analyte. To maintain a steady concentration of UVFs in the water, 50 L of freshly spiked water was renewed twice a day. Three fish were killed after 24, 48 and 72 h of exposure, and their muscle tissue was lyophilized and analyzed with the developed method to evaluate the residual analyte concentration.

Results and Discussion

Chromatographic Separation

To separate the UVFs, three columns were evaluated: an XDB C18, Eclipse C18, and SB C18. The first and second columns did not show good resolution of the critical pair AVO and EHMC (Rs < 0.8), and there was tailing in the avobenzone peak. The SB C18 column was able to separate all analytes with good peak shape and resolution (Fig. 1), even for the critical pair (Rs = 1.05), using an isocratic program without modifiers in the mobile phase. This is an advantage because in the literature, the use of gradient separation and modifiers in the mobile phase (cationic and anionic surfactants, cyclodextrin and EDTA) to separate UVFs has been reported [25, 31–33].

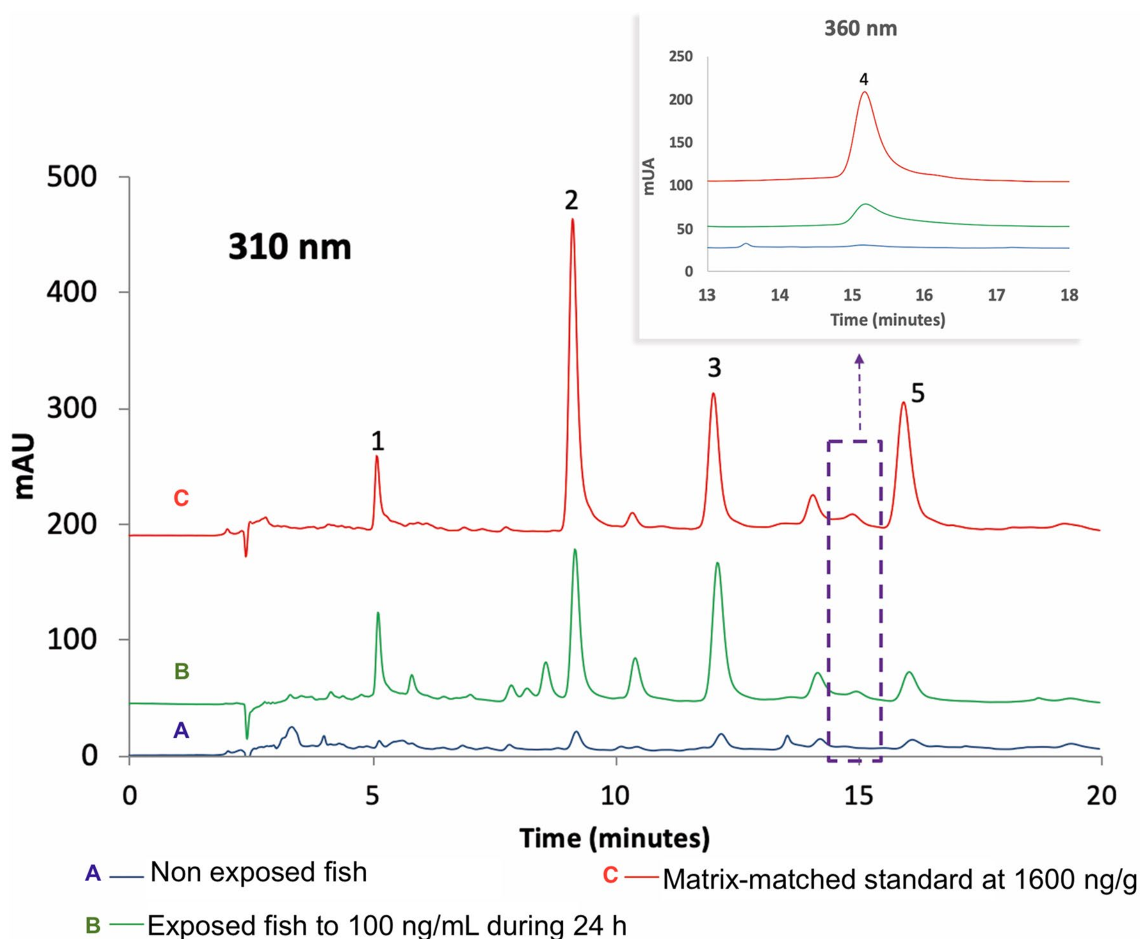

Fig. 1 Analysis of tilapia exposed for 24 h to UVF, matrix-matched standard at 1600 ng g^{-1} and non-exposed tilapia by off-line MSPD followed by on-line SPE–LC/UV method. Peaks: 1. oxybenzone, 2. MBC, 3. octocrylene, 4. avobenzone and 5. EHMC

Author's personal copy

Off-line MSPD Method Optimization

The MSPD conditions were optimized by maintaining a sample:C18 silica ratio of 1:2 and ACN as the elution solvent. Lipids are the main interferents present in biota extracts, and ACN was selected because of its low affinity for lipids [32]. To obtain a clear extract suitable for on-line SPE, two parameters were studied in the MSPD extraction: the use of a co-column packed into the bottom of the same column as the MSPD material, using 0.5 g of different sorbents (florisil, primary-secondary amine, and alumina) and the volume elution solvent (5 and 10 mL of ACN). The use of a co-column diminished the recoveries, so the optimal results were obtained without it. Additionally, 5 mL ACN was enough to recover more than 80% of most of the analytes (except EHMC) with minimum lipids in the extract. With 10 mL ACN, recoveries of EHMC were higher, but co-extracted matrix components (mainly lipids) generated an extract not suitable for on-line SPE.

Even with the above conditions, the extract obtained was turbid, so centrifugation (4000 rpm, 15 min) was applied for clarification.

On-line SPE–LC/UV Method Optimization

Because UVFs in biota are present at trace levels, most works report the use of LC–MS or GC–MS systems to improve sensitivity [8, 34]. In this work, on-line SPE preconcentration was employed to quantify UVFs at trace levels with the more affordable and common LC/UV system.

The high eluent strength of the extract recovered from MSPD reduced the retention of the analytes in the concentration column. To improve the retention of the compounds, 5 mL of the ACN extract was diluted to 10 mL (50% ACN), 15 mL (33% ACN) and 20 mL (25% ACN) with deionized water. The results are shown in Electronic Supplementary Material Fig. S2.

The use of 10 mL (50% ACN) caused breakthrough of oxybenzone and octocrylene from the precolumn. With 20 mL (25% ACN), the response of the most retained analytes (AVO and EHMC) decreased by between 30 and 40%; due to their low solubility in aqueous environments, they were possibly adsorbed to the centrifuge tubes. To test this hypothesis, the tubes were washed with 20 mL of 25% ACN and both analytes were detected. This UVFs behavior has been reported also with glassware [35].

The highest response for most analytes was achieved by dilution with 15 mL (33% ACN), even though the maximum response of oxybenzone was reduced by approximately 50%. Due to the different polarities of the analytes, developing a preconcentration process with high extraction yield for all of the analytes was very difficult. Thus, the maximum sensitivity was achieved for the most retained analytes (with the lowest response), even if the oxybenzone signal (with the highest response) was reduced.

Blank Contamination

It is difficult to monitor personal care products at trace levels because they are ubiquitous in the environment, and there are several contamination sources from daily products used by laboratory personnel, such as shampoos, soaps, lotions, cleaning agents and sun creams [12, 36]. Signals of UVFs were found in the control muscle, but they were < LOQs (Fig. 1). For this reason, during method validation and sample analysis from the exposure assay, interferences were routinely monitored by analyzing method blanks and samples from the control fish.

Matrix Effect

To evaluate the influence of the matrix on the preconcentration of the UVFs, the average peak area measured for the matrix-matched standard was compared with that obtained for the pure standard solution. The matrix-matched standard was prepared by adding 20 µL of a standard solution at 20 ng μL^{-1} (equivalent to 400 ng of each UVF) to 5 ml of an ACN extract obtained from control, lyophilized muscle by off-line MSPD. Then, 10 mL of deionized water was also added to obtain an ACN–water ratio of 33:67 v/v. On the other hand, a pure standard solution was prepared by adding the same quantity of UVFs to 15 mL of ACN–water 33:67 v/v. The solutions were prepared in triplicate and analyzed with the on-line SPE–LC/UV system.

A negative matrix effect with signal suppression was observed for octocrylene (− 4%) and EHMC (− 17%). On the other hand, a positive matrix effect was observed with enhancement of the signal for avobenzone (+ 13%), MBC (+ 14%) and oxybenzone (+ 85%). This significant positive effect in oxybenzone, the most polar analyte (log Kow = 3.5), could be explained by its higher retention in the concentration column caused by its interaction with matrix components. Because oxybenzone presented a strong matrix effect, recoveries for this UVF were obtained using a matrix-matched calibration curve to avoid quantitation errors. For the other analytes, a matrix effect between − 20 and + 20% was considered not significant [37]. The results are shown in Electronic Supplementary Material Fig. S3.

Method Validation

Table 2 shows the results of the method validation. Adequate linearity was found for all analytes from 100 to 3600 ng g^{-1}, obtaining r^2 values > 0.98. The accuracy was evaluated with recovery, and it was calculated from the slope of the equation obtained by plotting the amount recovered versus the

Author's personal copy

Table 2 Linearity, accuracy, precision, LODs and LOQs for the determination of UVF in fish muscle with the off-line MSPD method followed by on-line SPE–LC/UV

Parameter	Analyte				
	OXY	MBC	OCT	AVO	EHMC
Linearity (r^2) [a]	0.9901	0.9948	0.9905	0.9844	0.9867
Intercept[b]	−12 (±149)	−27 (±147)	−5 (±148)	−91 (±204)	−46 (±125)
Slope[b]	0.89 (±0.12)	0.85 (±0.12)	0.89 (±0.12)	0.96 (±0.17)	0.64 (±0.10)
Accuracy (recovery)[a]	89 (±12)	85 (±12)	89 (±12)	96 (±17)	64 (±10)
Precision (repeatability) 400 ng g^{-1} (RSD %, n=6)	19	14	24	16	22
Precision (repeatability) 1200 ng g^{-1} (RSD %, n=6)	8	15	2	20	11
LOD (S/N=3) (ng g^{-1})	8	12	16	7	13
LOQ (S/N=10) (ng g^{-1})	25	40	52	24	41

[a]Range of concentrations 100–3600 ng g^{-1}

[b]Confidence interval at 95%

amount added. Recoveries were between 85 and 96%, except for that of EHMC (64%). Precision was evaluated at two concentration levels (400 and 1200 ng g^{-1}), obtaining RSDs < 20% for all UVFs, except for octocrylene and EHMC spiked at 400 ng g^{-1}. The limits of detection (LODs) and quantitation (LOQs) were calculated at the optimum wavelength for each UVF, and they were between 7–16 ng g^{-1} and 24–52 ng g^{-1}, respectively. These limits were comparable to reported values [38].

Analysis of Incurred Samples

Incurred materials are useful samples for validation purposes. In these samples, the target analyte may be essentially extraneous, but it has been introduced before sampling. The contaminant is more closely bound in the matrix than in a spiked sample [39].

The validated off-line MSPD method followed by the on-line SPE–LC/UV method was used to measure the residual quantities of each UVF in the muscle of tilapia exposed for 3 days to 100 ng mL^{-1} of each analyte. Figure 1 illustrates typical chromatograms of the muscle samples from exposed tilapia and a matrix-matched standard.

The results showed that the five UV filters accumulated in fish muscle. In fish, the contaminant concentrations depend on the amounts of analyte in contact with the organism, the rate of uptake (through diet and respiration) and elimination (by metabolism, egestion, respiration, and growth) [6]. Figure 2 shows the concentrations of each analyte found in the incurred samples. The concentrations of MBC and

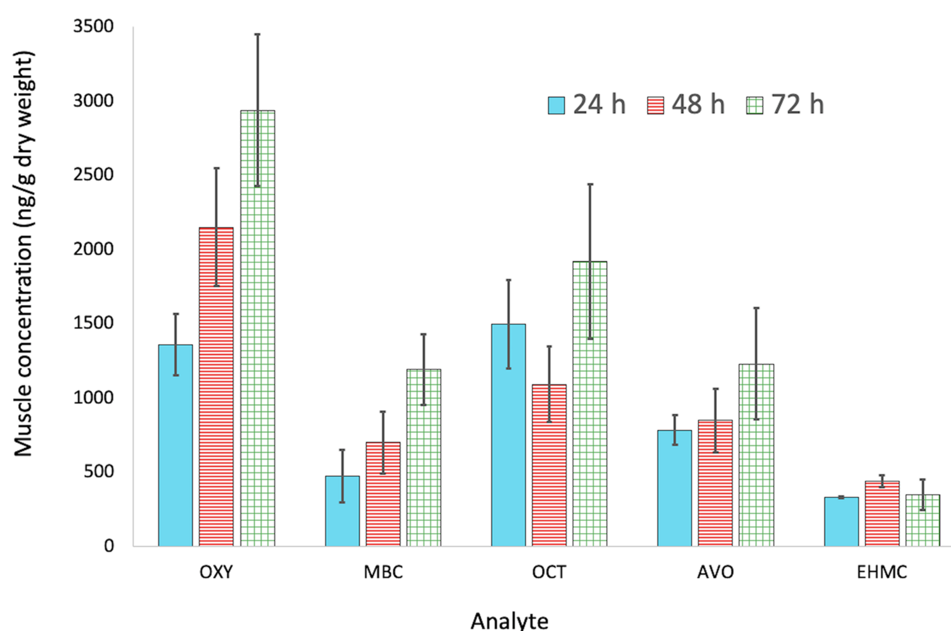

Fig. 2 Concentrations (ng g^{-1}) of UVF obtained from the analysis of exposed samples (n=3)

Author's personal copy

oxybenzone after 24 h were 540 and 1350 ng g^{-1} and increased to 1250 and 2900 ng g^{-1} after 72 h of exposure, respectively. The other compounds also increased in concentration from 24 to 72 h. For octocrylene, the concentrations were between 1200 and 1800 ng g^{-1}, for avobenzone, they were between 820 and 1300 ng g^{-1} and, finally, for EHMC, they were between 400 and 650 ng g^{-1}.

These results can be explained by the highest bioavailability of oxybenzone due to its hydrophilic character and relatively high water solubility (log Kow = 3.5; solubility = 13 mg L^{-1}) compared with those of the other UVFs studied, such as EHMC (log Kow = 5.8; solubility 0.15 mg L^{-1}). The lower accumulation of most hydrophobic UVFs could be justified by the low lipid content in tilapia muscle, which is approximately 2% w/w [40], because for many compounds, the bioaccumulation and log Kow are directly related, and the adipose tissue of organisms can accumulate hydrophobic analytes. However, this does not apply to hydrophilic analytes such as oxybenzone because they could appear in aqueous compartments, such as blood, generating a major incidence in tilapia [41].

Although there is scarce information about the behavior of UVF bioaccumulation in tilapia, these results agreed with the study reported by Chen et al. about the accumulation and elimination of synthetic musks and other kinds of personal care products (PCPs) in this kind of fish. They found that the most accumulated compounds were polar compounds, i.e., musk ketone (log Kow = 4.3) and musk xylene (log Kow = 4.8), in comparison with galaxolide and tonalide (log Kow = 5.7 and 5.9, respectively), which are more lipophilic [42].

Conclusions

A sensitive analytical method based on off-line MSPD followed by an on-line SPE–LC/UV method to determine five UVFs in tilapia muscle was developed. To the authors' knowledge, this method constitutes the first application of on-line SPE–LC to determine UVFs in fish muscle. The chromatographic separation was very simple, using an isocratic mode without any modifiers in the mobile phase. The method allowed for the quantification of UVFs at trace levels (ng g^{-1}) using a common and accessible LC/UV system. The matrix effect was only significant for oxybenzone, the most polar analyte. The method was applied effectively to analyze UVFs in tilapia that were artificially exposed to these contaminants. All of the analytes were found at levels between 400 and 2900 ng g^{-1}. The most accumulated analyte was oxybenzone, and the least accumulated analyte was EHMC after 72 h of exposure. The proposed method is a good alternative for environmental monitoring of UVFs in tilapia.

Acknowledgements This work was supported by Dirección General de Asuntos del Personal Académico from Universidad Nacional Autónoma de México (DGAPA-UNAM) projects PAPIIT: IA204717 and PAPIME: PE208418. Óscar E. Mogica-García thanks DGAPA-UNAM for the scholarship awarded. The authors want to thank Biologist Elena Loeza-Fuentes (Facultad de Medicina Veterinaria y Zootecnia, Departamento de Abejas, Conejos y Organismos Acuáticos, UNAM) for providing the tilapia and technical support.

The experimental procedures with tilapia were approved by the Bioethics Committee for Animal Health (CICUAL, Faculty of Chemistry, UNAM).

Compliance with ethical standards

Conflict of interest The authors declared no potential conflicts of interest with respect to the research, authorship and/or publication of this article.

References

1. Rainieri S, Barranco A, Primec M, Langerholc T (2017) Occurrence and toxicity of musks and UV filters in the marine environment. Food Chem Toxicol 104:57–68
2. Schneider SL, Lim HW (2019) Review of environmental effects of oxybenzone and other sunscreen active ingredients. J Am Acad Dermatol 80(1):266–271
3. Rastogi SC (2002) UV filters in sunscreen products—a survey. Contact Dermatitis 46:348–351
4. Chisvert A, Salvador A (2007) 31—UV filters in sunscreens and other cosmetics. Regulatory aspects and analytical methods. In: Salvador A, Chisvert A (eds) Analysis of cosmetic products. Elsevier, Amsterdam, pp 83–120
5. Volpe A, Pagano M, Mascolo G, Grenni P, Rossetti S (2017) Biodegradation of UV-filters in marine sediments. Sci Total Environ 575:448–457
6. Borgå KB (2013) Ecotoxicology: bioaccumulation. In: Encyclopedia of ecology. Elsevier. pp 346–348
7. Hassanin A, Kaminishi Y, Itakura T (2017) Characterization of Tilapia (*Oreochromis niloticus*) aldehyde reductase (AKR1A1) gene, promoter and expression pattern in benzo-a-pyrene exposed fish. Toxicol Mech Methods 27(1):36–44
8. Weisbrod CJ, Kunz PY, Zenker AK, Fent K (2007) Effects of the UV filter benzophenone-2 on reproduction in fish. Toxicol Appl Pharmacol 225:255–266
9. Montesdeoca-Esponda S, Checchini L, Del Bubba M, Sosa-Ferrera Z, Santana-Rodriguez JJ (2018) Analytical approaches for the determination of personal care products and evaluation of their occurrence in marine organisms. Sci Total Environ 633:405–425
10. Chisvert A, Benedé JL, Salvador A (2018) Current trends on the determination of organic UV filters in environmental water samples based on microextraction techniques—a review. Anal Chim Acta 1034:22–38
11. Chisvert A, Giokas D, Benedé J, Salvador A (2018) Environmental monitoring of cosmetic ingredients. In : Analysis of cosmetic products, 2nd edn, pp 435–547
12. Meinerling M, Daniels M (2006) A validated method for the determination of traces of UV filters in fish using LC–MS/MS. Anal Bioanal Chem 386:1465–1473
13. Tsai DY, Chen CL, Ding WH (2014) Optimization of matrix solid-phase dispersion for the rapid determination of salicylate and benzophenone-type UV absorbing substances in marketed fish. Food Chem 154:211–216

Author's personal copy

14. Barker SA (2007) Matrix solid phase dispersion (MSPD). J Biochem Biophys Methods 70:151–162
15. Capriotti AL, Cavaliere C, Foglia P, Samperi R, Stampachiacchiere S, Ventura S, Laganà A (2015) Recent advances and developments in matrix solid-phase dispersion. Trends Anal Chem 71:186–193
16. Fumes BH, Andrade MA, Franco MS, Lanças FM (2017) On-line approaches for the determination of residues and contaminants in complex samples. J Sep Sci 40:183–202
17. Olmos-Espejel JJ, García de Llasera MP, Velasco-Cruz M (2012) Extraction and analysis of polycyclic aromatic hydrocarbons and benzo[a]pyrene metabolites in microalgae cultures by off-line/on-line methodology based on matrix solid-phase dispersion, solid-phase extraction and high-performance liquid chromatography. J Chromatogr A 1262:138–147
18. Faraji M, Yamini Y, Gholami M (2019) Recent advances and trends in applications of solid-phase extraction techniques in food and environmental analysis. Chromatographia 82(8):1207–1249
19. Bony E, Mace Y, Pinto A, Delvaux D, Kiss R, Riant O, Feron O, Quetin-Leclercq J (2017) Validation of a SPE HPLC–UV method for the quantification of a new ER-specific photosensitizer OR-141 in blood serum using total error concept. J Pharm Biomed Anal 141:87–94
20. Kuklenyik P, Baker SE, Bishop AM, Morales AP, Calafat AM (2013) On-line solid phase extraction-high performance liquid chromatography–isotope dilution-tandem mass spectrometry approach to quantify *N*, *N*-diethyl-*m*-toluamide and oxidative metabolites in urine. Anal Chim Acta 787:267–273
21. Campone L, Piccinelli AL, Celano R, Pagano I, Russo M, Rastrelli L (2018) Rapid and automated on-line solid phase extraction HPLC–MS/MS with peak focusing for the determination of ochratoxin A in wine samples. Food Chem 244:128–135
22. Li J, Chen L, Wang X, Jin H, Ding L, Zhang K, Zhang H (2008) Determination of tetracyclines residues in honey by on-line solid-phase extraction high-performance liquid chromatography. Talanta 75:1245–1252
23. Liu R, Ruan T, Wang T, Song S, Guo F, Jiang G (2014) Determination of nine benzotriazole UV stabilizers in environmental water samples by automated on-line solid phase extraction coupled with high-performance liquid chromatography–tandem mass spectrometry. Talanta 120:158–166
24. Gago-Ferrero P, Mastroianni N, Díaz-Cruz MS, Barceló D (2013) Fully automated determination of nine ultraviolet filters and transformation products in natural waters and wastewaters by on-line solid phase extraction–liquid chromatography–tandem mass spectrometry. J Chromatogr A 1294:106–116
25. Oliveira HM, Segundo MA, Lima JLFC, Miró M, Cerdà V (2010) On-line renewable solid-phase extraction hyphenated to liquid chromatography for the determination of UV filters using bead injection and multisyringe-lab-on-valve approach. J Chromatogr A 1217(22):3575–3582
26. Camilleri J, Baudot R, Wiest L, Vulliet E, Cren-Olivé C, Daniele G (2015) Multiresidue fully automated online SPE-HPLC–MS/MS method for the quantification of endocrine-disrupting and pharmaceutical compounds at trace level in surface water. Int J Environ Anal Chem 95(1):67–81
27. Frederiksen H, Nielsen O, Skakkebaek NE, Juul A, Andersson AM (2017) UV filters analyzed by isotope diluted TurboFlow-LC–MS/MS in urine from Danish children and adolescents. Int J Hyg Environ Health Part A 220(2):244–253
28. Bury D, Brüning T, Koch HM (2019) Determination of metabolites of the UV filter 2-ethylhexyl salicylate in human urine by online-SPE–LC–MS/MS. J Chromatogr B 1110–1111:59–66
29. Pan J, Zhang C, Zhang Z, Li G (2014) Review of online coupling of sample preparation techniques with liquid chromatography. Anal Chim Acta 815:1–15
30. Gutiérrez-Valencia TM, García de Llasera MP (2017) On-line MSPD–SPE–HPLC/FLD analysis of polycyclic aromatic hydrocarbons in bovine tissues. Food Chem 223:82–88
31. Giokas DL, Sakkas VA, Albanis TA (2004) Determination of residues of UV filters in natural waters by solid-phase extraction coupled to liquid chromatography–photodiode array detection and gas chromatography–mass spectrometry. J Chrom A 1026(1–2):289–293
32. Chisvert A, Pascual-Martí M.C., Salvador A, (2001) Determination of the UV filters worldwide authorised in sunscreens by high-performance liquid chromatography: use of cyclodextrins as mobile phase modifier. J Chrom A 921(2):207–215
33. Schakel DJ, Kalsbeek D, Boer K (2004) Determination of sixteen UV filters in suncare formulations by high-performance liquid chromatography. J Chrom A 1049(1–2):127–130
34. Negreira N, Rodríguez I, Rodil R, Rubí E, Cela R (2013) Optimization of matrix solid-phase dispersion conditions for UV filters determination in biota samples. Int J Environ Anal Chem 93:1174–1188
35. Da Silva CP, Emídio ES, de Marchi MRR (2013) UV filters in water samples: experimental design on the SPE optimization followed by GC–MS/MS analysis. J Braz Chem Soc 24(9):1433–2144
36. Peck AM (2006) Analytical methods for the determination of persistent ingredients of personal care products in environmental matrices. Anal Bioanal Chem 386:907–939
37. EU Reference Laboratories (2019) Guidance document on analytical quality control and validation procedures for pesticide residues analysis in food and feed. https://www.eurl-pesticides.eu/library/docs/allcrl/AqcGuidance_Sanco_2013_12571.pdf. Accessed 25 Oct 2019.
38. Gago-Ferrero P, Díaz-Cruz MS, Barceló D (2012) An overview of UV-absorbing compounds (organic UV filters) in aquatic biota. Anal Bioanal Chem 404:2597
39. Magnusson B, Örnemark U (Ed.) (2014) Eurachem guide: the fitness for purpose of analytical methods—a laboratory guide to method validation and related topics. Second edition. EURACHEM https://www.eurachem.org/index.php/publications/guides/mv. Accessed 25 Oct 2019
40. Nutritionvalue.org (2019) Finfish and Shellfish Products. https://www.nutritionvalue.org/Fish%2C_raw%2C_tilapia_nutritional_value.html. Accessed 25 Oct 2019.
41. Gackowska A, Studziński W, Kudlek E, Dudziak M, Gaca J (2018) Estimation of physicochemical properties of 2-ethylhexyl-4-methoxycinnamate (EHMC) degradation products and their toxicological evaluation. Environ Sci Pollut Res Int 25:16037–16049
42. Chen G, Jiang R, Qiu J, Cai S, Zhu F, Ouyang G (2015) Environmental fates of synthetic musks in animal and plant: an in vivo study. Chemosphere 138:584–591

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.