



UNIVERSITÀ DI PARMA

ARCHIVIO DELLA RICERCA

University of Parma Research Repository

Ultra-Sensitive Solid-Phase Microextraction–Gas Chromatography–Mass Spectrometry determination of Polycyclic Aromatic Hydrocarbons in snow samples using a Deep Cavity BenzoQxCavitand

This is the peer reviewed version of the following article:

Original

Ultra-Sensitive Solid-Phase Microextraction–Gas Chromatography–Mass Spectrometry determination of Polycyclic Aromatic Hydrocarbons in snow samples using a Deep Cavity BenzoQxCavitand / Riboni, N.; Amorini, M.; Bianchi, F.; Pedrini, A.; Pinalli, R.; Dalcanale, E.; Careri, M.. - In: CHEMOSPHERE. - ISSN 0045-6535. - 303:(2022). [10.1016/j.chemosphere.2022.135144]

Availability:

This version is available at: 11381/2924328 since: 2024-04-27T15:53:27Z

Publisher:

Published

DOI:10.1016/j.chemosphere.2022.135144

Terms of use:

Anyone can freely access the full text of works made available as "Open Access". Works made available

Publisher copyright

note finali coverpage

(Article begins on next page)

**Ultra-Sensitive Solid-Phase Microextraction–Gas Chromatography–Mass Spectrometry
determination of Polycyclic Aromatic Hydrocarbons in snow samples using a Deep Cavity
BenzoQxCavitand**

N. Riboni^{1*}, M. Amorini¹, F. Bianchi^{1,2*}, A. Pedrini¹, R. Pinalli¹, E. Dalcanale¹, M. Careri¹

¹*University of Parma, Department of Chemistry, Life Sciences and Environmental Sustainability,
Parco Area delle Scienze 17/A, 43124 Parma, Italy*

²*University of Parma, Center for Energy and Environment (CIDEA), Parco Area delle Scienze 42,
43124 Parma, Italy*

Abstract

A very sensitive and selective solid-phase microextraction-gas chromatography-mass spectrometry method based on the use of a deep cavity BenzoQxCavitand as innovative coating was developed and validated for the simultaneous determination of the 16 US-EPA priority pollutants polycyclic aromatic hydrocarbons (PAHs) in snow samples at ultra-trace levels. The presence of an 8.3 Å deep hydrophobic cavity allowed the engulfment of all the 16 PAHs, providing enhanced selectivity also in presence of interfering aromatic pollutants at high concentration levels. Validation proved the reliability of the method for the determination of the investigated compounds achieving detection limits in the 0.03-0.30 ng/L range, good precision, with relative standard deviations <18% and recovery rates in the 90.8(±2.1)%-109.6(±1.0)%. The detection of low-molecular weight PAHs in snow samples from Antarctica and Alps confirms the

widespread occurrence of these compounds, thus assessing the impact of anthropogenic activities onto the environment.

Keywords: solid-phase microextraction, polycyclic aromatic hydrocarbons, priority pollutants, cavitand, trace analysis

Corresponding Authors

*Nicolò Riboni: e-mail: nicolo.riboni@unipr.it. Phone: +39 0521 905128

*Federica Bianchi: e-mail: federica.bianchi@unipr.it. Phone: +39 0521 905446

1. Introduction

In spite of the efforts made in the last decades for reducing environmental pollution, the release of compounds related to anthropogenic activities in different compartments is still an ongoing issue, considering the climate warming and ecosystems alteration. Among pollutants, great attention has been paid toward the monitoring of polycyclic aromatic hydrocarbons (PAHs) both to assess the impact of anthropogenic activities onto the environment and to study climate changing (Dat and Chang, 2017; Friedman et al., 2014; Manousi and Zachariadis, 2020; Szopińska et al., 2016; Yu et al., 2019). In fact, PAHs can be generated by natural phenomena like open burning and volcanic activities but their detection in the environment is mostly related to anthropogenic sources, primarily generated by the incomplete combustion of organic matter (industrial, residential and vehicular emissions), or due to petroleum spills (Abdel-Shafy and Mansour, 2016; Alegbeleye et al., 2017; Lawal, 2017; Mojiri et al., 2019). Currently, over 400 PAHs and derivatives have been identified, among which the compounds listed as priority pollutants by the United States Environmental Protection Agency (US-EPA) or the European Environment Agency, which have set strict regulations for the presence of these compounds in the environment (Keith, 2015; Lerda, 2010; Lestingi et al., 2017).

Recent studies demonstrated the occurrence of PAHs in different compartments, such as wastewater, groundwater, surface water, seawater, atmosphere and suspended particulate matter, in populated continents (Mojiri et al., 2019), polar regions-Arctic (Friedman et al., 2014; Yu et al., 2019) and Antarctica (Arcoleo et al., 2021, 2020; Potapowicz et al., 2019; Szopińska et al., 2016; Szumińska et al., 2021; Vecchiato et al., 2015) and high-altitude mountains (Arellano et al., 2018, 2014; Fernandez et al., 2021; Gabrieli et al., 2010; Guzzella et al., 2016), thus highlighting their environmental ubiquity. Being semi-volatile persistent organic pollutants (POPs), PAHs can be

easily absorbed onto the aerosol organic matter, leading to long-range atmospheric transport from the emission source to remote areas. Ambient temperature is one of the main parameters determining the deposition of semivolatile compounds: the drop of temperatures lead to the cold condensation of the aerodispersed organic matter and the deposition of the sorbed organic compounds, which are then accumulated in the snow deposits and cryosphere of high-altitude and high-latitude areas (Arellano et al., 2014; Cao et al., 2018; Szopińska et al., 2016; Vecchiato et al., 2018, 2015). Snowpacks can be therefore considered as temporary reservoirs of pollutants: seasonal melting leads to PAHs transfer to both soils and freshwater systems, becoming a serious treat to mountain and polar ecosystems. In addition, depending on temperature, these compounds can be transferred to deeper layers and preserved in glacial ice for long period or released during seasonal thawing. The increase of global temperatures related to climate warming is therefore a long-term hazard concern, being these compounds released from perennal ice and accumulated in biota (Szumińska et al., 2021). Besides the long-range transport, it was demonstrated that increased concentration levels of pollutants could be also associated with local sources of contamination related to human activities, such as Antarctica research stations (Arcoleo et al., 2021; Potapowicz et al., 2019; Szopińska et al., 2016; Szumińska et al., 2021; Vecchiato et al., 2015).

The analysis of PAHs levels is of pivotal importance in order to assess the impact of the anthropogenic activity on the local ecosystem, to study the long range transport mechanisms and for paleoclimatic studies. The major issues are related to the very low concentration present in these samples (in the ng/L level) and the lack of functional groups in the PAHs structure that can be effectively targeted for their binding. Therefore, proper sample pretreatment is required to preconcentrate the analytes and remove possible interfering compounds prior to the instrumental analysis (Jinadasa et al., 2020; Manousi and Zachariadis, 2020; Ncube et al., 2018; Raza et al.,

2018; Sajid et al., 2021). Solid phase extraction and liquid-liquid are the two conventional approaches for the extraction and preconcentration of the analytes from snow and water samples (Jinadasa et al., 2020; Manousi and Zachariadis, 2020; Ncube et al., 2018; Raza et al., 2018; Sajid et al., 2021). However, these techniques present several drawbacks, including time-consuming extraction steps, reduced automation, increased sample handling and large amounts of both sample and organic solvents. Being able to combine a significant reduction of sample volume, automation and minimal sample handling with no or very limited use of organic solvents, micro-extraction techniques have emerged (Manousi and Zachariadis, 2020; Ncube et al., 2018; Raza et al., 2018). Different miniaturized sample preparation techniques have been successfully applied for the extraction and preconcentration of PAHs from environmental matrices, including solid-phase microextraction (SPME), magnetic solid-phase extraction, stir bar sorptive extraction, micro-extraction in packed sorbent (MEPS) and dispersive liquid-liquid micro-extraction (Jinadasa et al., 2020; Manousi and Zachariadis, 2020; Ncube et al., 2018; Raza et al., 2018; Sajid et al., 2021). Since its development in 1997 (Pawliszyn, 1997), SPME has been considered one of the most intriguing microextraction techniques due to its simplicity, miniaturization, portability, automation, wide range of linear response and solvent-free approach. Currently, several commercial devices based on polymeric coatings are available, covering a wide range of polarity. However, being mostly based on aspecific hydrophobic and π - π interactions, commercial materials have very low selectivity toward a specific class of analytes, thus presenting limited enrichment capabilities. In order to overcome these limitations, novel sorbents have been proposed to be used for environmental monitoring, such as molecular imprinted polymers, ionic liquids, carbon nanotubes, metal organic frameworks and sol-gel based coatings (Delińska et al., 2021; Gómez - Ríos et al.2019; Murtada, 2020; Xu et al., 2013; Zheng et al., 2018).

In this context, the use of molecular receptors as SPME coatings is very attractive to address both selectivity and sensitivity issues: highly selective host molecules could be designed and developed to obtain specific host-guest interactions toward target analytes (Bertani et al., 2016; Bianchi et al., 2014, 2008, 2003; Gómez - Ríos et al., 2019; Li, 1999; Riboni et al., 2016; Trzciński et al., 2017). Our research group has already demonstrated the superior performances of designed quinoxaline cavitands for the extraction of aromatic analytes from environmental samples (Amorini et al., 2022; Bertani et al., 2016; Bianchi et al., 2014, 2008, 2003; Riboni et al., 2016; Trzciński et al., 2017). In the present study, a deep cavity cavitand, the BenzoQxCav is proposed as coating for the solid phase microextraction-gas chromatography-mass spectrometry (SPME-GC-MS) determination of the 16 US-EPA priority PAHs at trace levels in snow samples. After optimization by experimental design, the method was validated and applied for the analysis of snow samples from Antarctica and Italian Alps.

2. Experimental

2.1 Fiber preparation

BenzoQxCav was synthesized and characterized as reported in Amorini et al. (Amorini et al., 2022). Details regarding chemical and materials are reported in **S2.1**. The BenzoQxCav-based coatings were prepared by applying the dipping procedure already developed in our labs (Bianchi et al., 2014; Riboni et al., 2016): briefly, bare fused silica fibers were dipped into a hydrofluoric acid solution (40% v/v) for 10 s and immediately washed in Milli-Q water. The fiber coating was obtained by vertically dipping the activated silica fiber in the

Duralco 4460 epoxy glue and, after 2 min, in the BenzoQxCav powder (~40 mesh). Details regarding fiber morphology characterization and bleeding tests are reported in **S2.2**.

2.2 GC-MS analysis

Analytes were extracted by direct immersion of the developed SPME fiber in 9.5 ml sample solutions. The analysis was performed by gas chromatography-mass spectrometry (GC-MS), thermally desorbing the fiber directly in the GC-injector port at 300 °C. Internal standard method was used to improve method precision using the deuterated forms of 16-PAHs. Signals were acquired in time scheduled selected-ion monitoring mode (SIM), acquiring 3 ions per analytes, one used for quantitation and two for confirmatory purpose. Details regarding the GC-MS conditions are reported in supplementary data (**S2.3**).

2.4 Method optimization

The extraction conditions of the BenzoQxCav-coated fibers were optimized considering as main factors the extraction time and temperature performing a Box-Wilson central composite face centered design (CCF) to investigate the significant effects and applying the multicriteria method of the Derringer's desirability function approach to find the global optimal experimental conditions. Detailed information are reported in **S2.4**.

2.5 Method validation

Method validation was performed according to EURACHEM guidelines (Magnusson and Örnemark, 2014) under the optimized conditions using not contaminated tap water as blank matrix. Detection and quantitation limits were obtained based on the mean blank response and the standard

deviation of fortified blank matrix. Calibration curves were evaluated over three orders of magnitude, in the LOQ-100 ng/L range. Repeatability and batch-to-batch intermediate precision were evaluated as relative standard deviations (RSD%) of the responses of a blank fortified matrix at low, intermediate and high concentration levels. Trueness was calculated for each analyte in terms of recovery rate (RR%) at three concentration levels. Selectivity was evaluated by analyzing water samples containing 70 ng/L of each PAH spiked with a mixture of benzene, toluene, ethylbenzene and p- and o-xylene (BTEX) at the concentration of 1 µg/L each. The responses of PAHs in the BTEX-free and spiked water samples were compared by performing a Student t-test. Finally, the enrichment factors (EFs) of BenzoQxCav-coated fibers were obtained by analyzing a fortified matrix spiked with 25 ng/L of each PAH under the optimized extraction conditions. The results were compared with those of commercially available 30 µm PDMS fibers. Method validation additional information are reported in **S2.5**.

2.6 Analysis of surface snow samples from Antarctica and Italian Alps

The validated SPME-GC-MS method was finally applied for investigating the presence of PAHs in surface snow samples collected by the National Antarctic Research Program (PNRA) in the coastal area of Victoria Land (Antarctica) during the 2020–2021 austral summer. In particular, the analyzed snow samples were collected as represented in **Figure 1**: sample A nearby the meteorological station of *Icaro Camp* (74°42'37" S, 164°07'05" E); sample B in the area of the scientific research base *Enigma Lake* (74°42'38" S, 164°01'10" E); sample C north of Zucchelli station (74°36'55" S, 163°56'29" E); sample D near Cape Phillips (73°03'41" S, 169°36'16" E).

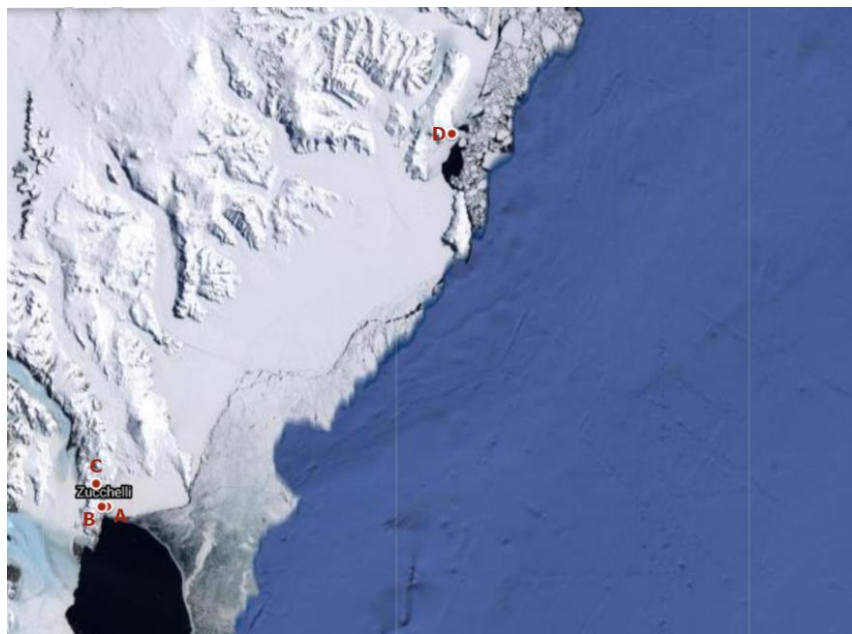


Figure 1. Sampling sites of surface snow in Antarctica.

Alpine snow samples were collected in the Dolomites alpine area in the following locations: sample 1 (46°25'13" N, 11°37'49" E), sample 2 (46°17'55" N, 11°48'16" E), sample 3 (46°19'29" N, 11°44'43" E), sample 4 (46°19'11" N, 11°48'12" E), sample 5 (46°23'41" N, 11°44'26" E) and sample 6 (46°23'0" N, 11°47'0" E) (**Figure 2**). Samples were taken in December 2021. Snow samples were manually sampled after removal of the first 10 centimeters of the snow to prevent contamination using solvent rinsed-glass bottles (100 mL volume). All bottles were rinsed twice with deionized water and dried prior sampling.

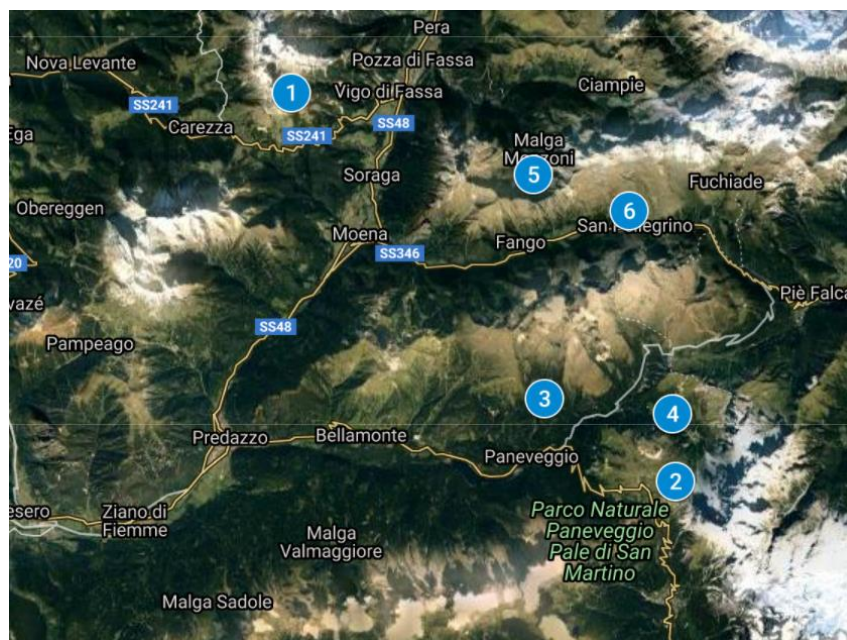


Figure 2. Sampling sites of surface snow in Dolomites.

Samples were delivered and kept at -20 °C until analysis, thawed under chemical hood in an organic-free environment at ambient temperature in dark boxes in order to prevent solar irradiation. The samples were then spiked with the ISs and submitted to analysis. Three replicate measurements per sample were performed.

3. Results and discussion

Starting from the QxCav receptor (Bianchi et al., 2008, 2003), quinoxaline cavitands have demonstrated superior capabilities for the extraction of aromatic guests, being able to bind target molecules by hydrophobic, π - π and CH- π interactions. The enhanced selectivity can be achieved by exploiting specific interactions with functional groups of the target molecule (Bianchi et al., 2014), promoting its confinement inside the cavity (Bertani et al., 2016) and/or by

conformationally blocking the cavity to strengthen the interactions between the host and the guest (Riboni et al., 2016; Trzciński et al., 2017).

Since PAHs do not present functional groups that can be effectively targeted for specific binding, BenzoQxCav (**Figure 3**) was synthesized to maximize π - π and CH- π interactions of the analytes with the cavity walls by creating a 2.5 Å deeper cavity compared to the QxCav precursor (Amorini et al., 2022).

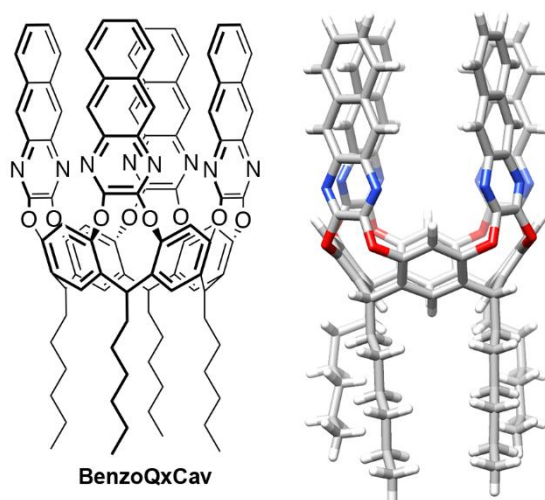


Figure 3. Left: structure of the BenzoQxCav; right: BenzoQxCav molecular structure

In the present study, the inclusion capabilities of the BenzoQxCav were exploited to develop a SPME coating for trace determination of the 16 US-EPA priority pollutants in snow samples. The previously demonstrated high thermal resistance of BenzoQxCav (Amorini et al., 2022) is required to allow the thermal desorption of the analytes in the GC injector port. Coating thickness and morphology were investigated by means of SEM under different magnifications: a homogeneous, porous and uniform coating (**Figure S1**) was observed with an average thickness in the 17-22(\pm 4) μ m range (batch of 3 fibers, 3 measured points per fiber). In order to evaluate the background

content of PAHs in the coating, the fibers were desorbed at 300 °C, demonstrating the absence of the target analytes in the sorbent material itself.

3.1 SPME Optimization

Based on previous knowledge and the instrumental limitation, the experimental domain was studied as follows: i) extraction time in the 15-45 min range, considering 15 min as the minimum time for an effective adsorption of the analytes and 45 as maximum time to avoid prolonged analysis time; ii) extraction temperature ranging between 40 and 80 °C: 40 °C were selected to obtain a stable thermostating, whereas 80 °C was selected as maximum temperature to avoid both the desorption of the analytes from the coating and the increase of the pressure inside the vials.

Salt addition was not optimized since it is known that a reduction of fiber lifetime can occur when immersion analyses are performed, due to the formation of crystals onto the fiber.

For each analyte, the significance of linear, quadratic and interaction effects was evaluated using a forward search step-wise variable algorithm (p to remove 0.05). The obtained models are shown in **Table S1**.

The response surfaces for fluorene, anthracene, fluoranthene, pyrene, benzo[*b*]fluoranthene and dibenzo[*a,h*]anthracene are represented in **Figure S2**. As shown, no common trend of the response surfaces is present for the different PAHs, therefore a multicriteria method for identifying the global optimal extraction conditions is required.

By applying the Derringer's desirability function approach, the optimal global extraction conditions resulted in an extraction temperature of 80 °C and an extraction time of 45 min. A global desirability value $D=0.81$ and single desirability values in the 0.67-0.93 range were calculated, thus providing a good extraction of all PAHs. The achieved findings proved that a

global response enhancement was obtained when a long extraction time was applied; as for the temperature, an opposite behavior was observed for the low- and high-molecular weight PAHs, respectively, being the low-molecular weight compounds more easily desorbed by the fiber coating than the heaviest ones. These findings can be rationalized considering that a temperature rise is able to increase the overall extraction kinetic, enhancing the analyte diffusion coefficients (Xu et al., 2020; Zhang et al., 2018) and the BenzoQxCav breathing, thus facilitating the intake of the bulky analytes.

3.5 Method validation

The validation of the developed SPME-GC-MS method allowed to achieve LODs and LOQs values obtaining values in the 0.03-0.30 and 0.09-1.01 ng/L range, respectively. The achieved LODs were better or comparable with the lowest limits reported in previously published studies related to the extraction of PAHs using different extraction techniques and coating materials (Jalili et al., 2020; Jinadasa et al., 2020; Ncube et al., 2018; Raza et al., 2018; Sajid et al., 2021). Additional advantages rely on the reduced sample handling with a limited sample volume, thus minimizing the risk of contamination and reducing the analytical variability. Method linearity was demonstrated for all PAHs over three order of magnitude (**Table 1**).

266 **Table 1:** LODs, LOQs and regression coefficients for the 16 US-EPA PAHs.

Analyte	LODs (ng/L)	LOQs (ng/L)	Regression coefficient* b(±s _b)
Naphthalene	0.09	0.29	0.1408(±0.0054)
Acenaphthylene	0.21	0.71	0.0984(±0.0042)
Acenaphthene	0.24	0.80	0.0962(±0.0027)
Fluorene	0.07	0.23	0.0670(±0.0017)
Phenanthrene	0.03	0.09	0.0684(±0.0020)
Anthracene	0.07	0.23	0.0662(±0.0026)
Fluoranthene	0.03	0.09	0.0636(±0.0019)
Pyrene	0.03	0.09	0.0716(±0.0021)
Benzo[<i>a</i>]anthracene	0.13	0.42	0.0641(±0.0023)
Chrysene	0.09	0.31	0.0622(±0.0015)
Benzo[<i>b</i>]fluoranthene	0.10	0.34	0.0664(±0.0018)
Benzo[<i>k</i>]fluoranthene	0.11	0.38	0.0872(±0.0035)
Benzo[<i>a</i>]pyrene	0.30	1.01	0.0773(±0.0021)
Indeno[1,2,3- <i>c,d</i>]pyrene	0.14	0.47	0.0697(±0.0023)
Dibenzo[<i>a,h</i>]anthracene	0.10	0.34	0.0506(±0.0010)
Benzo[<i>g,h,i</i>]perylene	0.17	0.57	0.0831(±0.0022)

*Calibration curve: y=bx

268
269 As for method precision, both repeatability and intermediate precision provided excellent results
270 with RSDs always lower than 18% (**Table S2**). ANOVA was applied to evaluate the presence of

significant differences among the 3 days of analysis used for evaluating intermediate precision, thus highlighting that mean values were not significantly different (p values > 0.05). Excellent results were also found in terms of trueness, obtaining recovery rates in the 90.8(\pm 2.1)-109.6(\pm 1.0)% range, thus assessing the efficiency of the developed SPME-GC-MS method (**Table S2**).

Finally, the selectivity of the devised method was investigated by extracting the target analytes in presence of a mixture of BTEX having a concentration 14 times higher than that of PAHs. The results were then compared with those achieved by extracting the PAHs in a blank fortified matrix without BTEX. As illustrated in **Figure S3**, no significant difference was observed between the GC responses, thus demonstrating the high selectivity of the developed coating. In fact, despite the high concentration of the interfering compounds and their possible interaction adsorption via hydrophobic, π - π and CH- π interaction, the BenzoQxCav coating was characterized by a very high selectivity towards the investigated analytes. This behavior can be rationalized by recalling that the number of π - π and CH- π interactions of the analytes with the cavity walls is responsible of the overall binding. This number is higher for PAHs than for BTEX in the case of the deep cavitand BenzoQxCav, since the interaction with the resorcinarene moiety at the bottom of the cavity is essential to promote the binding (Vincenti and Dalcanale, 1995). Regarding the selectivity of the material toward PAHs in the presence of other hydrophobic compounds of environmental concern, like polychlorinated biphenyl compounds, it is possible to hypothesize an interaction with the supramolecular receptor mainly via π - π interactions; by contrast, as for nitro-PAHs, it can be hypothesized that nitro derivatives are less retained due to the lower affinity for the cavity of the nitro groups and to the additional bulkiness related to the introduction of nitro groups in PAH.

The extraction performance of the BenzoQxCav-coated fibers was investigated in terms of enrichment factors (EFs). The EFs of the developed fibers were in the 10260(\pm 340)-125500(\pm 4300)

range and resulted to be generally higher than those obtained by using the PDMS fiber, which varied in the 1420(\pm 42)-43800(\pm 4400) range. As for the extraction mechanism, it has to be considered that an adsorption mechanism is present in the case of the BenzoQxCav due to the presence of a solid porous coating, whereas an absorption mechanism is associated to the use of PDMS fibers, being the analytes mainly extracted by partitioning into a “liquid-like” phase. Finally, the stability of the BenzoQxCav-coated fiber was demonstrated by performing over 150 analyses, without a significant loss in the extraction performance.

3.7 Analysis of Antarctica and Alpine snow samples

Considering the excellent performance in terms of sensitivity, the SPME-GC-MS method based on the BenzoQxCav coated fiber was applied for the analysis of surface snow samples from both Antarctica and Alps. PAHs in the surface snow samples showed total concentrations (Σ 16PAHs) in the 32.2-49.5 ng/L range (**Table 2**). According to already published results, PAHs were detected in all samples, thus demonstrating the widespread presence of these compounds in the Antarctica region (Arcoletto et al., 2021, 2020; Potapowicz et al., 2019; Szumińska et al., 2021; Vecchiato et al., 2015). Naphthalene was detected in all the investigated samples with concentration levels ranging from 8.11(\pm 0.21) to 12.36(\pm 0.98) ng/L, accounting for the 20-37% of the total PAHs concentration. Three- and four-ring PAHs consisted of 62-78% of the Σ 16PAHs; by contrast, the five- and six-rings PAHs were present in very low amounts in only 2 samples out of 4. Benzo[a]anthracene was detected only in samples A and B, respectively, thus accounting for the 8.2 and 16% of the Σ 16PAHs present in the two sampled sites.

As previously reported, the presence of high levels of low-molecular weight compounds, namely naphthalene, acenaphthene, fluorene and phenanthrene, could be related to fuel combustion from

both local and long range sources (Arcoleo et al., 2021; Szumińska et al., 2021; Vecchiato et al., 2015). Regarding high-molecular weight PAHs, considering that these compounds are less prone to long-range transportation (Szumińska et al., 2021), their presence can be ascribed to local combustion phenomena. Sample A was characterized by the highest levels of pollution among the investigated samples, with ultra-trace amounts of benzo[*a*]pyrene, indeno[1,2,3-*c,d*]pyrene, dibenzo[*a,h*]anthracene and benzo[*g,h,i*]perylene. This sample was collected nearby the meteorological station of *Icaro Camp*, which is close to the Zucchelli research station. Therefore the presence of the heaviest PAHs could be ascribed to local combustion sources (Arcoleo et al., 2021; Szumińska et al., 2021; Vecchiato et al., 2015). Compared to the results published by Arcoleo et al. (Arcoleo et al., 2021), who analyzed samples collected in the same areas, higher levels of $\Sigma 16$ PAHs were obtained. Although the concentrations of the low-weight analytes were very similar, the proposed method being characterized by LOQs lower than those of the MEPS-GC-MS method, allowed a more sensitive detection of five and six-rings PAHs.

340

341 **Table 2:** Concentration (ng/L) of the 16 monitored US-EPA PAHs in the Antarctica snow samples

	Sample A	Sample B	Sample C	Sample D
Naphthalene	12.36(±0.98)	8.56(±0.34)	11.89(±0.30)	8.11(±0.21)
Acenaphthylene	1.05(±0.03)	n.d.	n.d.	3.59(±0.12)
Acenaphthene	1.09(±0.06)	1.38(±0.04)	0.96(±0.06)	1.22(±0.06)
Fluorene	10.18 (±0.13)	7.90(±0.48)	6.72(±0.38)	8.26 (±0.38)
Phenanthrene	2.04(±0.05)	2.19(±0.03)	1.34(±0.04)	1.71(±0.06)
Anthracene	0.66(±0.02)	n.d.	n.d.	n.d.
Fluoranthene	5.61(±0.29)	6.26(±0.32)	4.91(±0.25)	5.35(±0.31)
Pyrene	4.21(±0.29)	5.64(±0.38)	5.89(±0.28)	4.09(±0.19)
Benzo[<i>a</i>]anthracene	4.06(±0.13)	6.77(±0.30)	n.q.	n.d.
Chrysene	2.59(±0.06)	3.18(±0.09)	n.d.	n.d.
Benzo[<i>b</i>]fluoranthene	n.d.	n.d.	n.d.	n.d.
Benzo[<i>k</i>]fluoranthene	n.d.	n.d.	n.d.	n.d.
Benzo[<i>a</i>]pyrene	2.21(±0.32)	n.d.	n.d.	n.d.
Indeno[1,2,3- <i>c,d</i>]pyrene	1.56(±0.06)	n.q.	n.q.	1.13(±0.02)
Dibenzo[<i>a,h</i>]anthracene	n.d.	n.d.	n.d.	n.d.
Benzo[<i>g,h,i</i>]perylene	1.89(±0.11)	n.q.	n.q.	1.16(±0.03)

342 n.d.: not detected (<LOD)

343 n.q.: not quantitated (<LOQ)

344

The method was then applied for the analysis of surface snow samples collected at high altitude in the Italian Alpine area (over 2000 m, except for Sample 4, which was sampled at 1824 m). All the sampling sites could be reached only by foot and were located far from traffic areas except for Sample 6, which is located nearby an alpine pass. The obtained results are showed in **Table 3**.

Table 3: Concentration (ng/L) of the 16 monitored US-EPA PAHs in the Alpine snow samples

	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
Naphthalene	4.17(±0.19)	5.62(±0.38)	3.87(±0.19)	12.7(±0.69)	8.86(±0.33)	16.6(±0.71)
Acenaphthylene	n.d.	0.84(±0.03)	n.d.	1.12(±0.04)	n.q.	2.21(±0.05)
Acenaphthene	n.q.	n.d.	n.q.	0.96(±0.05)	n.q.	2.52(±0.11)
Fluorene	1.74(±0.04)	1.12(±0.06)	0.94(±0.02)	1.96(±0.11)	2.07(±0.05)	3.32(±0.11)
Phenanthrene	2.21(±0.09)	2.08(±0.04)	1.73(±0.05)	6.67(±0.23)	4.10(±0.22)	8.83(±0.12)
Anthracene	0.27(±0.02)	n.d.	n.q.	0.35(±0.01)	0.48(±0.02)	0.41(±0.01)
Fluoranthene	2.88(±0.04)	3.02(±0.12)	1.97(±0.15)	9.88(±0.09)	5.26(±0.16)	8.20(±0.23)
Pyrene	1.96(±0.04)	2.54(±0.09)	1.68(±0.03)	6.01(±0.45)	3.81(±0.10)	5.54(±0.13)
Benzo[a]anthracene	n.q.	0.52(±0.03)	n.d.	0.98(±0.03)	0.62(±0.01)	0.72(±0.01)
Chrysene	0.54(±0.04)	0.38(±0.01)	n.d.	0.74(±0.04)	1.76(±0.05)	2.74(±0.10)
Benzo[b]fluoranthene	0.44(±0.03)	n.q.	n.d.	2.69(±0.11)	0.98(±0.06)	2.27(±0.05)
Benzo[k]fluoranthene	0.47(±0.01)	n.d.	n.d.	0.54(±0.01)	n.q.	0.97(±0.01)
Benzo[a]pyrene	n.d.	n.q.	n.d.	n.q.	n.d.	1.31(±0.07)
Indeno[1,2,3-c,d]pyrene	n.d.	0.48(±0.02)	0.52(±0.02)	1.24(±0.02)	0.68(±0.05)	1.56(±0.12)
Dibenzo[a,h]anthracene	n.d.	n.d.	n.d.	1.15(±0.04)	n.d.	0.88(±0.08)
Benzo[g,h,i]perylene	n.d.	n.q.	n.d.	0.65(±0.02)	n.d.	1.05(±0.08)

n.d.: not detected (<LOD)

n.q.: not quantitated (<LOQ)

The obtained results are in agreement with the findings reported in literature by Arellano et al. (Arellano et al., 2018, 2014) and Gabrieli et al. (Gabrieli et al., 2010) regarding the concentrations of PAHs in snow samples collected in the alpine areas. As previously reported, the compound with the highest concentration is naphthalene, which was detected in all samples in the 3.87(±0.19)-16.6(±0.71) ng/L range, thus accounting for 26.1-38.0% of the Σ16PAHs. Fluorene, phenanthrene, fluoranthene and pyrene were also present in all the analyzed samples, with concentration levels in the 0.94(±0.02)-3.32(±0.11), 1.73(±0.04)-8.83(±0.12), 1.97(±0.15)-9.88(±0.09) and 1.68(±0.03)-6.01(±0.45) ranges, respectively. As shown in the table, low-molecular weight PAHs accounted for more than 85% of the Σ16PAHs, in accordance with previously reported findings (Arellano et al., 2018, 2014; Gabrieli et al., 2010). The prevalence of three- and four-rings PAHs in high altitude snowpacks have been reported in Europe, North America and Asia (Arellano et al., 2018). This behavior can be explained taking into account that snow is considered a very efficient scavenger of PAHs present in the atmosphere both in the gas phase and absorbed onto particulate matter. Pollutants can be transported to high elevation, where the temperature drop enhances the condensation of these compounds and the particulate matter acts as core for the nucleation of aqueous crystals, thus entrapping PAHs in the snowflakes. In addition, the porous structure and high surface/volume ratio of snowflakes increase the scavenging capabilities, capturing aerosol particles and gaseous hydrophobic molecules (Gabrieli et al., 2010). As reported in **Table 4**, the highest concentration of the Σ16PAHs was detected in Sample 6: these findings could be related to the presence of anthropic activities due to the presence of tourists. Finally, the concentrations obtained by applying the developed method were compared with those assessed by using the

EPA8272 method, properly adapted for the detection of high molecular weight PAHs. Non-significant differences between the results were achieved for the majority of the compounds (Tables S3 and S4). Since the LOQs of the reference method were higher than those presented in this study for most of the investigated PAHs, the quantitation of the analytes present at sub-ng/L level could not be feasible by using the reference method. However, the matching of all the quantifiable concentrations proved the reliability of the developed method.

4. Conclusions

In the present study, a new SPME coating based on the BenzoQxCav receptor was developed and tested for the SPME-GC-MS determination of PAHs at trace levels. The developed material provided excellent performance in terms of extraction capabilities and selectivity, thus resulting in very low detection and quantitation limits. The high affinity of the coating towards PAHs was related to the complexation of the target analytes inside the receptor *via* π - π and CH- π interactions. The enhanced selectivity was possible due to the presence of a 8.3 Å deep cavity, suitable for hosting bulky guests such as PAHs, with a very high degree of fitting. The extraction capabilities of the selective coating were not influenced by the presence of other potential interfering aromatic compounds also when present at concentration levels 14 times higher than those of the investigated PAHs.

The analysis of snow samples from Antarctica and Alps confirmed the widespread presence of low-molecular weight PAHs.

The major features of the proposed method are related to the high sensitivity and selectivity, the reduced sample handling and the possibility of matching the requirements of green analytical

chemistry. Therefore, it will be used in future studies to screen the presence of PAHs in different environment to assess the anthropogenic impact and to study pollutant transport mechanisms.

Acknowledgements

This work has benefited from the equipment and framework of the COMP-HUB Initiative, funded by the ‘Departments of Excellence’ program of the Italian Ministry for Education, University and Research (MIUR, 2018–2022).

The study was also supported by the project “Emerging COntaminants in Antarctic Snow: sources and TRANsport (ECO AS:TRA)”, funded by the MIUR Programma Nazionale di Ricerca in Antartide (PNRA), grant: PNRA18_00229.

Author Contributions

Conceptualization: F. Bianchi, N. Riboni, E. Dalcanale, R. Pinalli; Data analysis: F. Bianchi, N. Riboni, M. Amorini. Investigation: F. Bianchi, N. Riboni, M. Amorini, R. Pinalli, A. Pedrini. Methodology: F. Bianchi, N. Riboni, E. Dalcanale, M. Careri. Writing – original draft: F. Bianchi, N. Riboni. Funding acquisition: F. Bianchi, M. Careri. All authors edited and reviewed the manuscript.

References

- Abdel-Shafy, H.I., Mansour, M.S.M., 2016. A review on polycyclic aromatic hydrocarbons: source, environmental impact, effect on human health and remediation. *Egypt. J. Pet.* 25, 107–123. <https://doi.org/10.1016/j.ejpe.2015.03.011>
- Alegbeleye, O.O., Opeolu, B.O., Jackson, V.A., 2017. Polycyclic aromatic hydrocarbons: a critical

review of environmental occurrence and bioremediation. *Environ. Manage.* 60, 758–783.

<https://doi.org/10.1007/s00267-017-0896-2>

Amorini, M., Riboni, N., Pesenti, L., Dini, V.A., Pedrini, A., Massera, C., Gualandi, C., Bianchi, F., Pinalli, R., Dalcanale, E., 2022. Reusable cavitand-based electrospun membranes for the removal of polycyclic aromatic hydrocarbons from water. *Small* 2104946, 1–8. <https://doi.org/10.1002/sml.202104946>

Arcoleo, A., Bianchi, F., Careri, M., 2021. A sensitive microextraction by packed sorbent-gas chromatography-mass spectrometry method for the assessment of polycyclic aromatic hydrocarbons contamination in Antarctic surface snow. *Chemosphere* 282, 131082. <https://doi.org/10.1016/j.chemosphere.2021.131082>

Arcoleo, A., Bianchi, F., Careri, M., 2020. Helical multi-walled carbon nanotube-coated fibers for solid-phase microextraction determination of polycyclic aromatic hydrocarbons at ultra-trace levels in ice and snow samples. *J. Chromatogr. A* 1631, 461589. <https://doi.org/10.1016/j.chroma.2020.461589>

Arellano, L., Fernández, P., Van Drooge, B.L., Rose, N.L., Nickus, U., Thies, H., Stuchlík, E., Camarero, L., Catalan, J., Grimalt, J.O., 2018. Drivers of atmospheric deposition of polycyclic aromatic hydrocarbons at European high-altitude sites. *Atmos. Chem. Phys.* 18, 16081–16097. <https://doi.org/10.5194/acp-18-16081-2018>

Arellano, L., Grimalt, J.O., Fernández, P., Lopez, J.F., Nickus, U., Thies, H., 2014. Persistent organic pollutant accumulation in seasonal snow along an altitudinal gradient in the Tyrolean Alps. *Environ. Sci. Pollut. Res.* 21, 12638–12650. <https://doi.org/10.1007/s11356-014-3196->

x

Bertani, F., Riboni, N., Bianchi, F., Brancatelli, G., Sterner, E.S., Pinalli, R., Geremia, S., Swager,

T.M., Dalcanale, E., 2016. Triptycene-roofed quinoxaline cavitands for the supramolecular detection of BTEX in air. *Chem. - A Eur. J.* 22, 3312–3319. <https://doi.org/10.1002/chem.201504229>

Bianchi, F., Bedini, A., Riboni, N., Pinalli, R., Gregori, A., Sidisky, L., Dalcanale, E., Careri, M., 2014. Cavitand-based solid-phase microextraction coating for the selective detection of nitroaromatic explosives in air and soil. *Anal. Chem.* 86, 10646–10652. <https://doi.org/10.1021/ac5025045>

Bianchi, F., Mattarozzi, M., Betti, P., Bisceglie, F., Careri, M., Mangia, A., Sidisky, L., Ongarato, S., Dalcanale, E., 2008. Innovative cavitand-based sol-gel coatings for the environmental monitoring of benzene and chlorobenzenes via solid-phase microextraction. *Anal. Chem.* 80, 6423–6430. <https://doi.org/10.1021/ac800881g>

Bianchi, F., Pinalli, R., Ugozzoli, F., Spera, S., Careri, M., Dalcanale, E., 2003. Cavitands as superior sorbents for benzene detection at trace level. *New J. Chem.* 27, 502–509. <https://doi.org/10.1039/b210942e>

Cao, S., Na, G., Li, R., Ge, L., Gao, H., Jin, S., Hou, C., Gao, Y., Zhang, Z., 2018. Fate and deposition of polycyclic aromatic hydrocarbons in the Bransfield Strait, Antarctica. *Mar. Pollut. Bull.* 137, 533–541. <https://doi.org/10.1016/j.marpolbul.2018.10.045>

Dat, N.D., Chang, M.B., 2017. Review on characteristics of PAHs in atmosphere, anthropogenic sources and control technologies. *Sci. Total Environ.* 609, 682–693. <https://doi.org/10.1016/j.scitotenv.2017.07.204>

Delińska, K., Rakowska, P.W., Kloskowski, A., 2021. Porous material-based sorbent coatings in solid-phase microextraction technique: recent trends and future perspectives. *TrAC - Trends Anal. Chem.* 143. <https://doi.org/10.1016/j.trac.2021.116386>

- Fernandez, P., van Drooge, B.L., Arellano, L., Grimalt, J.O., 2021. Atmospheric deposition of semivolatile organic pollutants in European high mountains: sources, settling and chemical degradation. *Sci. Total Environ.* 784, 147099. <https://doi.org/10.1016/j.scitotenv.2021.147099>
- Friedman, C.L., Zhang, Y., Selin, N.E., 2014. Climate change and emissions impacts on atmospheric PAH transport to the Arctic. *Environ. Sci. Technol.* 48, 429–437. <https://doi.org/10.1021/es403098w>
- Gabrieli, J., Decet, F., Luchetta, A., Valt, M., Pastore, P., Barbante, C., 2010. Occurrence of PAH in the seasonal snowpack of the Eastern Italian Alps. *Environ. Pollut.* 158, 3130–3137. <https://doi.org/10.1016/j.envpol.2010.06.042>
- Gómez-Ríos, G.A., Reyes Garcés, N., Tascon, M., 2019. Smart Materials in Solid Phase Microextraction (SPME). *Handbook of Smart Materials in Analytical Chemistry.* 581–619. <https://doi.org/10.1002/9781119422587.ch18>
- Guzzella, L., Salerno, F., Freppaz, M., Roscioli, C., Pisanello, F., Poma, G., 2016. POP and PAH contamination in the southern slopes of Mt. Everest (Himalaya, Nepal): long-range atmospheric transport, glacier shrinkage, or local impact of tourism? *Sci. Total Environ.* 544, 382–390. <https://doi.org/10.1016/j.scitotenv.2015.11.118>
- Jalili, V., Barkhordari, A., Ghiasvand, A., 2020. Solid-phase microextraction technique for sampling and preconcentration of polycyclic aromatic hydrocarbons: a review. *Microchem. J.* 157, 104967. <https://doi.org/10.1016/j.microc.2020.104967>
- Jinadasa, B.K.K.K., Monteau, F., Morais, S., 2020. Critical review of micro-extraction techniques used in the determination of polycyclic aromatic hydrocarbons in biological, environmental and food samples. *Food Addit. Contam. - Part A Chem.* 37, 1004–1026.

<https://doi.org/10.1080/19440049.2020.1733103>

Keith, L.H., 2015. The source of U.S. EPA's sixteen PAH priority pollutants. *Polycycl. Aromat. Compd.* 35, 147–160. <https://doi.org/10.1080/10406638.2014.892886>

Lawal, A.T., 2017. Polycyclic aromatic hydrocarbons. A review. *Cogent Environ. Sci.* 3. <https://doi.org/10.1080/23311843.2017.1339841>

Lerda, D., 2010. Polycyclic aromatic hydrocarbons (PAHs) Factsheet. JRC Tech. Notes 3, 1–25.

Lestingi, C., Tavaloni, T., Bardeggia, V., Perugini, M., Piersanti, A., 2017. A fit-for-purpose method to monitor 16 European Union PAHs in food: results of five years of official food control in two Italian regions. *Food Addit. Contam. - Part A Chem.* 34, 1140–1152. <https://doi.org/10.1080/19440049.2017.1325969>

Li, S., 1999. Artificial receptor-facilitated solid-phase microextraction of barbiturates. *Anal. Chem.* 71, 2146–2151. <https://doi.org/10.1021/ac980587o>

Magnusson, B., Örnemark, U., 2014. Eurachem Guide: The Fitness for Purpose of Analytical Methods – A Laboratory Guide to Method Validation and Related Topics, (2nd ed. 2014), Eurachem. <https://doi.org/978-91-87461-59-0>

Manousi, N., Zachariadis, G.A., 2020. Recent advances in the extraction of polycyclic aromatic hydrocarbons from environmental samples. *Molecules* 25, 1–29. <https://doi.org/10.3390/molecules25092182>

Mojiri, A., Zhou, J.L., Ohashi, A., Ozaki, N., Kindaichi, T., 2019. Comprehensive review of polycyclic aromatic hydrocarbons in water sources, their effects and treatments. *Sci. Total Environ.* 696, 133971. <https://doi.org/10.1016/j.scitotenv.2019.133971>

Murtada, K., 2020. Trends in nanomaterial-based solid-phase microextraction with a focus on environmental applications - A review. *Trends Environ. Anal. Chem.* 25, e00077.

<https://doi.org/10.1016/j.teac.2019.e00077>

Ncube, S., Madikizela, L., Cukrowska, E., Chimuka, L., 2018. Recent advances in the adsorbents for isolation of polycyclic aromatic hydrocarbons (PAHs) from environmental sample solutions. *TrAC - Trends Anal. Chem.* 99, 101–116.
<https://doi.org/10.1016/j.trac.2017.12.007>

Pawliszyn, J., 1997. *Solid Phase Microextraction: Theory and Practice*, Wiley–VCH, New York. Wiley-VCH.

Potapowicz, J., Szumińska, D., Szopińska, M., Polkowska, Ż., 2019. The influence of global climate change on the environmental fate of anthropogenic pollution released from the permafrost: Part I. Case study of Antarctica. *Sci. Total Environ.* 651, 1534–1548.
<https://doi.org/10.1016/j.scitotenv.2018.09.168>

Raza, N., Hashemi, B., Kim, K.H., Lee, S.H., Deep, A., 2018. Aromatic hydrocarbons in air, water, and soil: sampling and pretreatment techniques. *TrAC - Trends Anal. Chem.* 103, 56–73.
<https://doi.org/10.1016/j.trac.2018.03.012>

Riboni, N., Trzcinski, J.W., Bianchi, F., Massera, C., Pinalli, R., Sidisky, L., Dalcanale, E., Careri, M., 2016. Conformationally blocked quinoxaline cavitand as solid-phase microextraction coating for the selective detection of BTEX in air. *Anal. Chim. Acta* 905, 79–84.
<https://doi.org/10.1016/j.aca.2015.12.005>

Sajid, M., Nazal, M.K., Ihsanullah, I., 2021. Novel materials for dispersive (micro) solid-phase extraction of polycyclic aromatic hydrocarbons in environmental water samples: a review. *Anal. Chim. Acta* 1141, 246–262. <https://doi.org/10.1016/j.aca.2020.07.064>

Szopińska, M., Namieśnik, J., Polkowska, Ż., 2016. How Important Is Research on Pollution Levels in Antarctica? Historical Approach, Difficulties and Current Trends, in: *Reviews of*

535 Environmental Contamination and Toxicology. 79–156.
 536 https://doi.org/10.1007/398_2015_5008
 537 Szumińska, D., Potapowicz, J., Szopińska, M., Czapiewski, S., Falk, U., Frankowski, M.,
 538 Polkowska, Ż., 2021. Sources and composition of chemical pollution in Maritime Antarctica
 539 (King George Island), part 2: organic and inorganic chemicals in snow cover at the Warszawa
 540 Icefield. *Sci. Total Environ.* 796. <https://doi.org/10.1016/j.scitotenv.2021.149054>
 541 Trzciniński, J.W., Pinalli, R., Riboni, N., Pedrini, A., Bianchi, F., Zampolli, S., Elmi, I., Massera,
 542 C., Ugozzoli, F., Dalcanale, E., 2017. In search of the ultimate benzene sensor: the EtQxBox
 543 solution. *ACS Sensors* 2, 590–598. <https://doi.org/10.1021/acssensors.7b00110>
 544 Vecchiato, M., Argiriadis, E., Zambon, S., Barbante, C., Toscano, G., Gambaro, A., Piazza, R.,
 545 2015. Persistent organic pollutants (POPs) in Antarctica: occurrence in continental and
 546 coastal surface snow. *Microchem. J.* 119, 75–82.
 547 <https://doi.org/10.1016/j.microc.2014.10.010>
 548 Vecchiato, M., Barbaro, E., Spolaor, A., Burgay, F., Barbante, C., Piazza, R., Gambaro, A., 2018.
 549 Fragrances and PAHs in snow and seawater of Ny-Ålesund (Svalbard): local and long-range
 550 contamination. *Environ. Pollut.* 242, 1740–1747.
 551 <https://doi.org/10.1016/j.envpol.2018.07.095>
 552 Vincenti, M., Dalcanale, E., 1995. Host–guest complexation in the gas phase. Investigation of the
 553 mechanism of interaction between cavitands and neutral guest molecules. *J. Chem. Soc.,*
 554 *Perkin Trans. 2* 0, 1069–1076. <https://doi.org/10.1039/P29950001069>
 555 Xu, J., Hu, Q., Liu, X., Wei, S., Zheng, J., Lin, W., Ye, Y., Zhu, F., Ouyang, G., 2020.
 556 Determination of the mass transfer coefficients in direct immersion solid-phase
 557 microextraction. *J. Sep. Sci.* jssc.201901345. <https://doi.org/10.1002/jssc.201901345>

558 Xu, J., Zheng, J., Tian, J., Zhu, F., Zeng, F., Su, C., Ouyang, G., 2013. New materials in solid-
559 phase microextraction. *TrAC Trends Anal. Chem.* 47, 68–83.
560 <https://doi.org/10.1016/J.TRAC.2013.02.012>

561 Yu, Y., Katsoyiannis, A., Bohlin-Nizzetto, P., Brorström-Lundén, E., Ma, J., Zhao, Y., Wu, Z.,
562 Tych, W., Mindham, D., Sverko, E., Barresi, E., Dryfhout-Clark, H., Fellin, P., Hung, H.,
563 2019. Polycyclic aromatic hydrocarbons not declining in Arctic air despite global emission
564 reduction. *Environ. Sci. Technol.* 53, 2375–2382. <https://doi.org/10.1021/acs.est.8b05353>

565 Zhang, L., Gionfriddo, E., Acquaro, V., Pawliszyn, J., 2018. Direct immersion solid-phase
566 microextraction analysis of multi-class contaminants in edible seaweeds by gas
567 chromatography-mass spectrometry. *Anal. Chim. Acta* 1031, 83–97.
568 <https://doi.org/10.1016/j.aca.2018.05.066>

569 Zheng, J., Huang, J., Yang, Q., Ni, C., Xie, X., Shi, Y., Sun, J., Zhu, F., Ouyang, G., 2018.
570 Fabrications of novel solid phase microextraction fiber coatings based on new materials for
571 high enrichment capability. *TrAC - Trends Anal. Chem.* 108, 135–153.
572 <https://doi.org/10.1016/j.trac.2018.08.021>

573