Supplementary Information

Solvent-free determination of selected polycyclic aromatic hydrocarbons in plant material used for food supplements preparation: optimization of a solid phase microextraction method

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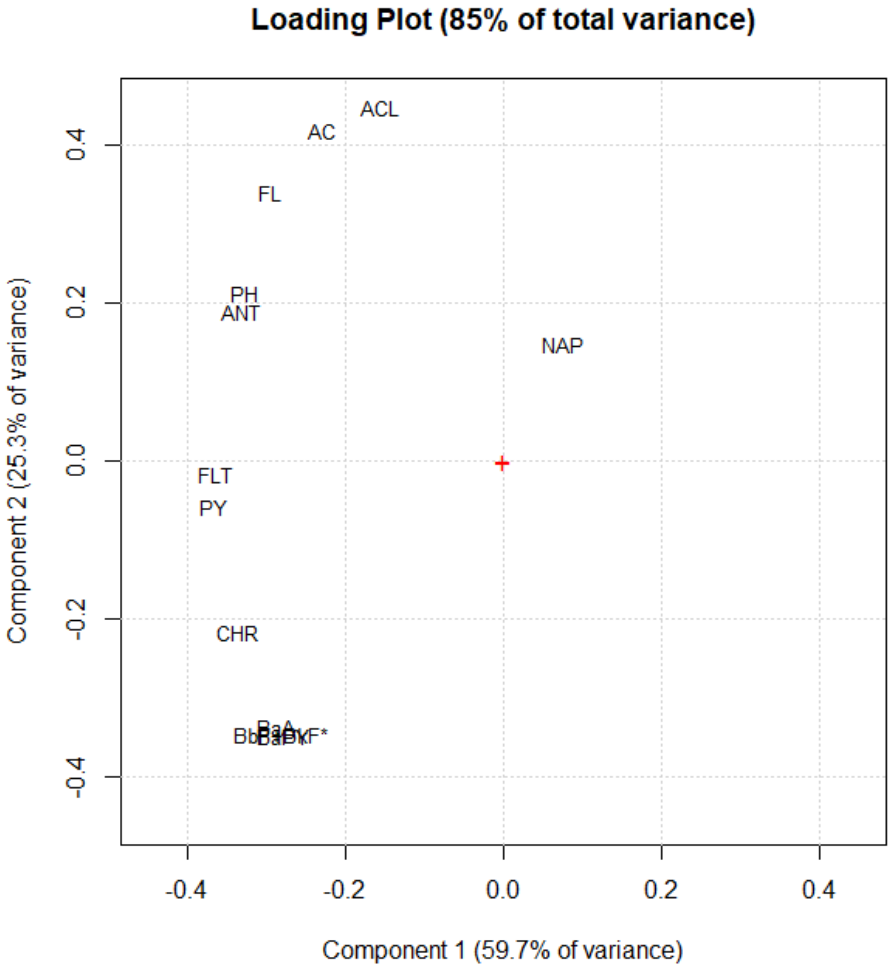


Figure S1. Loading plot of the PCA, performed on the areas obtained by the experiments of the first experimental design. The similar loadings on PC1 indicate a correlation among most analytes.

Table S1. Optimal values of the studied variables to obtain maximum sensitivity for light and heavy PAHs (MM: 128-202 Da and 228-278 Da, respectively).

Variable	Optimal value for L-PAHs	Optimal value for H-PAHs
t_{EXP}	50 min	90 min
T_{EXP}	70 °C	100 °C
t_{INC}	20 min	20 min
t_{DES}	3 min	3 min

Table S2. Selected ions for SIM detection and retention times of the 16 PAHs under study as well as the internal standards.

Compound	Selected ion in SIM mode (m/z)	Retention time (min)	Assigned internal standard
Analytes			
Napthatlene (NAP)	128	9.11	d-NAP
acenaphthylene (ACL)	152	12.84	d-AC
acenaphthene (AC)	154	13.26	d-AC
fluorene (FL)	166	14.50	d-PH
phenanthrene (PH)	178	16.76	d-PH
anthracene (ANT)	178	16.89	d-PH
fluoranthene (FLT)	202	19.60	d-PH
pyrene (PY)	202	20.12	d-PH
benzo[a]anthracene (BaA)	228	23.00	d-PH
chrysene (CHR)	228	23.08	d-PH
benzo[b]fluoranthene (BbF)*	252	25.66	d-PH

benzo[k]fluoranthene (BkF)*	252	25.74	d-PH
benzo[a]pyrene (BaPY)	252	26.55	d-PH
indeno[1,2,3-c,d]pyrene (IcdPY)	276	30.58	d-PE
dibenzo[a,h]anthracene (DahA)	278	30.72	d-PE
benzo[g,h,i]perylene (BghiPE)	276	31.37	d-PE
Internal standards			
d-Napthatlene (d-NAP)	136	9.06	
d-acenaphthene (d-AC)	164	13.19	
d-phenanthrene (d-PH)	188	16.71	
d-perylene (d-PE)	264	26.74	

* Benzo[b]fluoranthene and benzo[k]fluoranthene are considered as a unique peak (partially coeluted).

Table S3. Comparison of the present method with others in the literature, in terms of the main figures of merit and method characteristics.

Reference	**LOQ** (ng g^{-1})	**Precision** (RSD)	**Trueness**	**Sample amount** (g)	**Sample preparation**
Present work	3.1-14.7	3.4-20.9 %	88-105 %	0.1	SPME
Orecchio et al. (Ref 9)	0.2-0.5	10-20 %	76-102 %	5	SLE[a]
Ratola et al. (Ref 42)	0.7-2.3	5.8-8.2 %	72-100 %	10	SLE
De Nicola et al. (Ref 12)	2.6-8	4-15 %	≈ 70%	5	SLE
De Nicola et al. (Ref 43)	0.1-3.4	2.4-12 %	85-143 %	0.25	MSPD[b]

a- Solid-Liquid Extraction
b- Matrix Solid Phase Dispersion

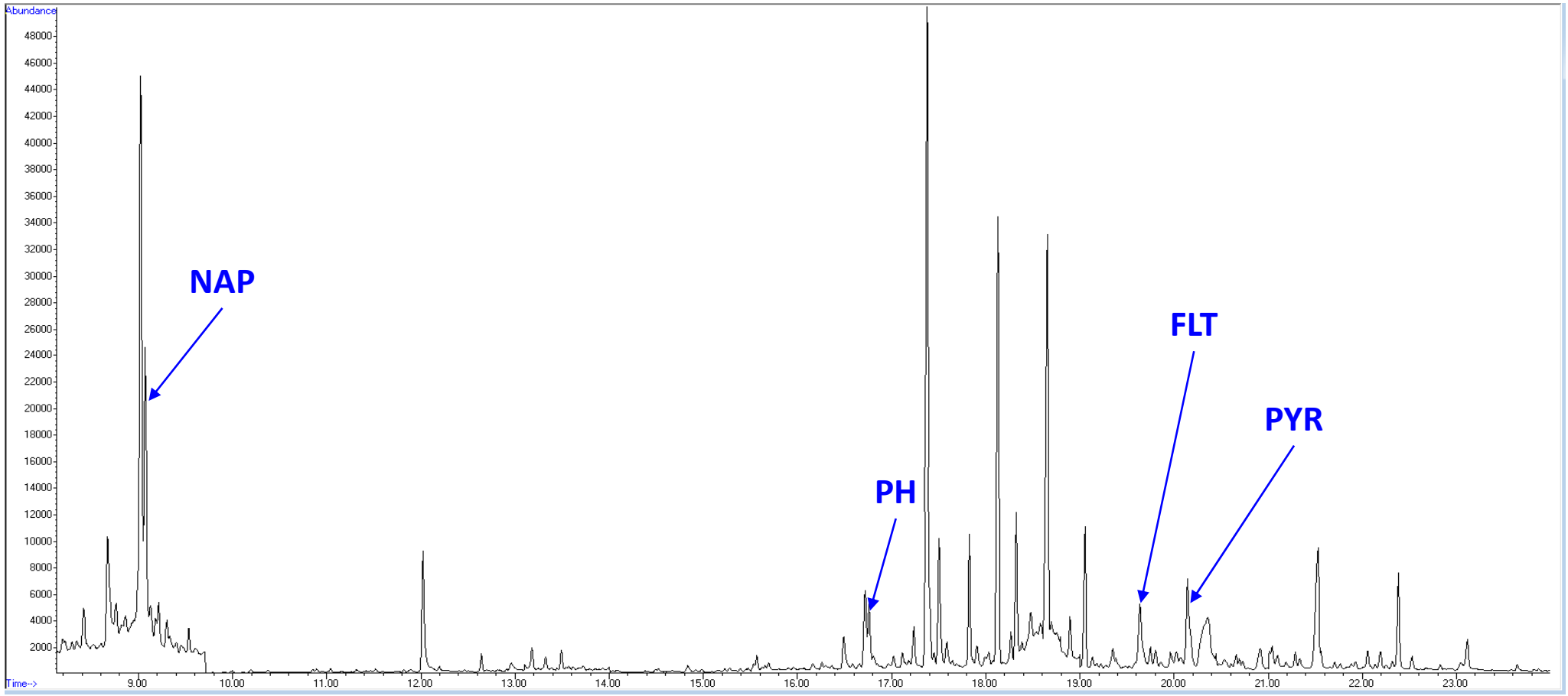


Figure S2: GC-MS chromatogram in Total Ion Current (TIC) of sample S3. The most concentrated analytes are indicated.

Table S4. Results of the quantitative analysis, expressed as ng g^{-1} of fresh buds' samples ± standard deviation (SD) (n=4).

Analyte	Sample S1	Sample S2	Sample S3
		ng g^{-1} ± SD	
NAP	5 ± 1	7.3 ± 0.9	8 ± 2
ACL	< LOD	< LOD	< LOD
AC	3.7 ± 0.7	4 ± 1	4.0 ± 0.5
FL	*3.4 ± 0.3 **	*3.4 ± 0.7 **	*3.5 ± 0.5 **
PH	9.3 ± 0.2	21 ± 3	15 ± 2
ANT	< LOD	5 ± 0.6	4.8 ± 0.4
FLT	10 ± 2	20 ± 4	16 ± 3
PY	7 ± 1	42 ± 4	21 ± 5
BaA	15 ± 2	17 ± 2	16 ± 1
CHR	6.1 ± 0.4	9 ± 3	6.1 ± 0.8
BbF+ BkF	< LOD	< LOD	< LOD

*Values at the LOQ level

Table S5. Experiments performed within the first experimental design; real values of each variable are reported.

Experiment	X_1	X_2	X_3	X_4
	T_{EXP} (°C)	t_{INC} (min)	t_{EXP} (min)	t_{DES} (min)
1	40	20	20	1
2	70	20	20	1
3	40	50	20	1
4	70	50	20	1
5	40	20	50	1
6	70	20	50	1
7	40	50	50	1
8	70	50	50	1
9	40	20	20	5
10	70	20	20	5
11	40	50	20	5
12	70	50	20	5
13	40	20	50	5
14	70	20	50	5
15	40	50	50	5
16	70	50	50	5
17	40	35	35	3
18	70	35	35	3

19	55	20	35	3
20	55	50	35	3
21	55	35	20	3
22	55	35	50	3
23	55	35	35	1
24	55	35	35	5
25	55	35	35	3
26	55	35	35	3
27	55	35	35	3

Table S6. Experiments performed within the second experimental design; real values of each variable are reported.

Experiment	$\mathbf{X_1}$	$\mathbf{X_3}$
	T_{EXP} (°C)	t_{EXP} (min)
1	80	60
2	100	60
3	80	90
4	100	90
5	80	75
6	100	75
7	90	60
8	90	90
9	90	75
10	90	75
11	90	75