RESTEK Pure Chromatography

Analysis of furan and alkylfurans in food commodities using headspace SPME arrow and GC-MS

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Introduction

- Furan and alkylfurans are produced during heating of foodstuff as a result of the thermal degradation of compounds such as carbohydrates, ascorbic acid, and derivatives, as well as some lipids.
- The International Agency for Research of Cancer classified furan as a possible carcinogenic compound, and there is a general concern on the possible health risks associated with the occurrence of furans and alkylfurans in food.
- Methods reported for the analysis of these volatile organic compounds include static headspace (HS) and solid phase microextraction (SPME) in combination with GC-MS.
- The use of SPME for the analysis of these highly volatile analytes has demonstrated improved method sensitivity and higher S/N for some of the alkylfurans. However, the fragility of traditional SPME fibers can be a concern.
- In this work, we present a HS-SPME-GC-MS method for the analysis of furans and alkylfurans in baby formula and coffee using a SPME arrow.

Goal

To develop a workflow using the SPME arrow coupled to GC-MS for the analysis of furan and alkylfurans in baby formula and coffee

Methods

Instrumental Analysis

Table 1. GC-MS conditions for the analysis of furan and alkylfurans.

Instrument	Agilent 7890B GC & 5977B MSD
Column	Rxi-624Sil MS, 30 m, 0.25 mm ID, 1.40 µm (Restek cat.# 13868)
Injection Mode	Split (1:10 and 1:100)
Liner	Topaz 1.8 mm ID SPME/straight liner (cat# 23280)
Inj. Temp.	280°C
Split Flow	14.0 mL/min (10:1) and 140 mL/min (100:1)
Purge Flow	5 mL/min
Oven	35°C (hold 3 min) to 75°C by 8°C/min, then to 200°C (hold 1 min) by 25°C/min
Carrier Gas	He, constant flow
Flow Rate	1.40 mL/min
Analyzer	MS (quadrupole)
Acquisition Type	SIM
Ionization Mode	EI (70 eV)
Transfer Line Temp.	280 °C
Source Temp.	325 °C
Quadrupole Temp.	200 °C
Solvent delay	2.2 min

Table 2. MS parameters for the analysis of furan, alkylfurans, and their internal standards (SIM mode).

Segment start time, min	Compound (Rt, min)	Ions	Dwell time, ms
2.2	Furan (2.447)	39	50
		68*	
	Furan-d4 (2.428)	42	
		72*	
4.2	2-Methylfuran (4.536)	53	30
		81	
		82*	
	3-Methylfuran (4.846)	53	30
		81	
		82*	
	2-Methylfuran-d6 (4.464)	58	30
		88*	
6.6	2-Ethylfuran (7.100)	53	30
		81*	
		96	
	2-Ethylfuran-d5 (7.001)	55	30
		101*	
	2,5-Dimethylfuran (7.243)	67	30
		95*	
	2,5-Dimethylfuran-d3 (7.179)	84	30
		99*	
10.6	2-Pentylfuran (11.570)	81	30
		138*	30
	2-Pentylfuran-d11 (11.501)	83	30
		149*	30

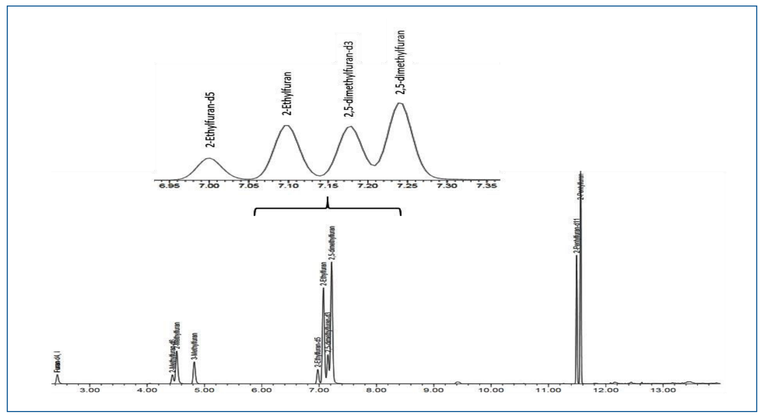

Figure 1. GC-MS chromatogram corresponding to furan and alkyl furan standards and internal standards.

Sample Preparation

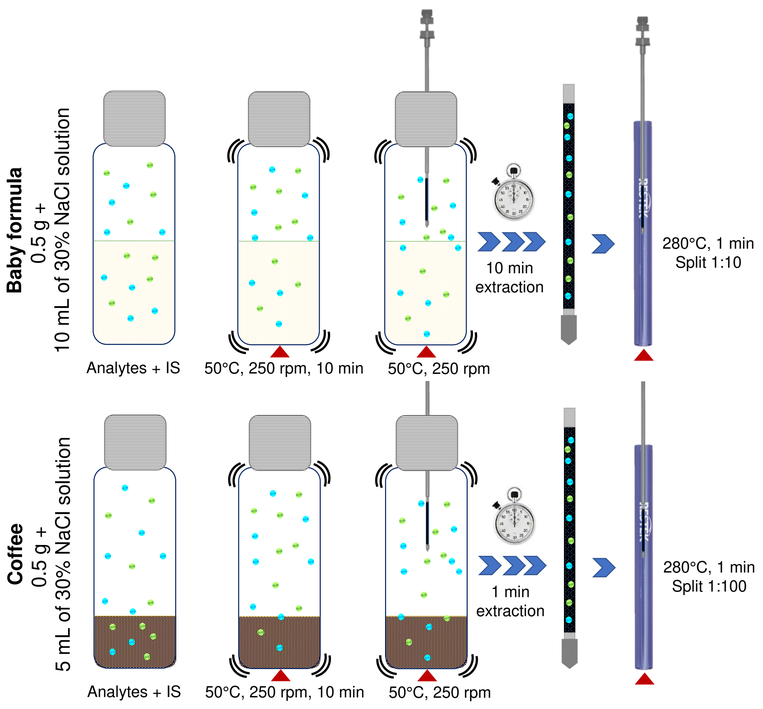

Figure 2. Sample preparation workflow.

Table 3. Sample preparation conditions for baby formula and coffee samples.

Sample	Baby formula	Coffee
Amount	0.5 g	
Volume of 30% NaCl solution added	10 mL	5 mL
Volume of internal standard solution added	50 µL (1 µg/mL solution)	40 µL (25 µg/mL solution)
Incubation time	10 min	
Incubation and extraction temperature	50°C	
Agitation	250 rpm	
HS-SPME extraction time	10 min	1 min
Desorption temperature	280°C	
Desorption time	1 min	

Calibration

- Calibration curves were constructed for all the target analytes in sodium chloride solution (30%) using appropriate deuterated analogues for each of them.
- Two calibration curves were prepared in 30% sodium chloride solution one for the analysis of low concentrations of furans in baby formula (1.25 – 150 ng in vial), and one for the quantitation of high concentrations target analytes in coffee (25 – 8000 ng in vial).
- Internal standard solutions were added to each calibration vial (50 µL of the 1 µg/mL solution for the low concentration calibration curve, and 40 µL of the 25 µg/mL solution for the high concentration calibration curve).
- Calibration curves were then constructed by plotting analyte area/ISTD average area ratios (n=2) versus spiked concentration.

Results and Discussion

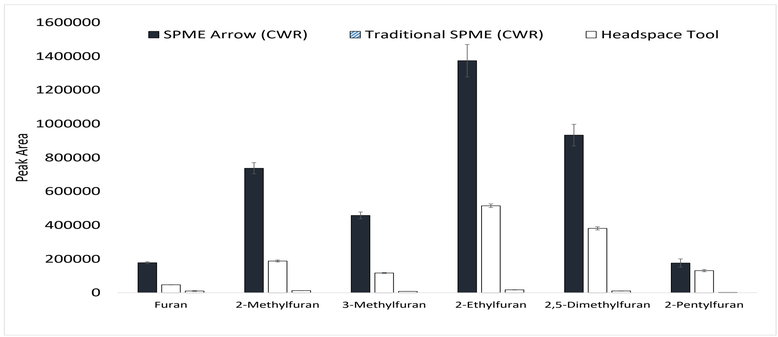

Figure 3. Responses of all analytes obtained with three sample prep techniques (n=3). CWR: Carbon Wide Range.

- **Comparison of sample prep techniques:** we performed a comparison among HS-SPME Arrow, HS-SPME (traditional fiber), and HS (Figure 3). For the SPME devices, we selected a CWR coating because it is the best phase for the extraction of highly volatile compounds. The HS-SPME-based techniques significantly outperformed HS analysis, and HS-SPME Arrow provided the best results in terms of analyte responses in all the cases. Based on this, we chose HS-SPME arrow.
- **Desorption conditions**: Based on a carryover evaluation, desorption temperature and time of 280°C and 1 min, respectively, were selected.
- **Extraction temperature:** The effect of three extraction temperatures (40, 50 and 60°C) was evaluated on the responses of analytes spiked in baby formula. Results demonstrated a decrease in the response of the most volatile analytes with an increase of the extraction temperature. 2-ethylfuran and 2,5-dimethylfuran did not show a significant difference in the responses obtained at 40 and 50°C, whereas 2-pentylfuran exhibited a slight increase in the response with an increase in the extraction temperature. We considered 50°C to be a good compromise for all the analytes. 10 min was selected as incubation time.
- **Extraction time:** Extraction time profiles were constructed for all target analytes. Extractions were carried out from the headspace of vials containing baby formula and sodium chloride solution spiked with all the analytes at 40 µg/kg. Extractions were conducted for 1, 2, 5, 10 and 20 min. As shown in Figure 4, the most volatile analytes (e.g. furan) reached equilibrium at 10 min, whereas the rest of the compounds did not reach a plateau at the evaluated extractions times. As a compromise, 10 min was chosen as the extraction time.

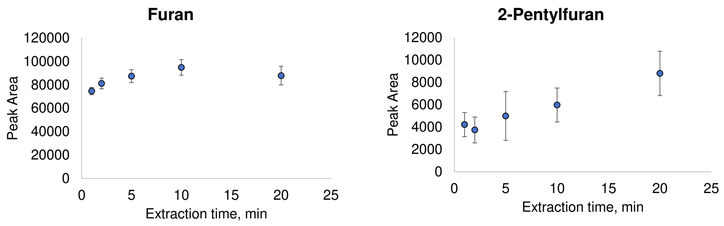

Figure 4. Representative extraction time profiles (n=3).

- **Coffee analysis**: To enable the analysis of coffee with the SPME arrows, the GC split was set to 1:100; the extraction time was set to 1 min; and the volume of sodium chloride solution was adjusted to 5 mL.

Table 4. Analysis of furan and alkylfurans in baby formula (top) and instant coffee (bottom).

Analyte	Blank (n=3)		Low conc.(n=3), 5 µg/kg*		High conc. (n=3), 50 µg/kg*	
	µg/kg	RSD, %	Acc., %	RSD, %	Acc., %	RSD, %
Furan	16	1	110	2	94	4
2-Methylfuran	60	2	97	2	100	3
3-Methylfuran	-	-	113	3	107	6
2-Ethylfuran	67	2	93	3	105	5
2,5-Dimethylfuran	-	-	101	4	97	14
2-Pentylfuran	219	3	87	11	108	12
Analyte	**Blank (n=3)**		**Low conc.(n=3), 1000 µg/kg***		**High conc. (n=3), 4000 µg/kg***	
	µg/kg	RSD, %	Acc., %	RSD, %	Acc., %	RSD, %
Furan	394	10	87	2	125	3
2-Methylfuran	843	10	94	3	116	2
3-Methylfuran	96	10	87	2	119	3
2-Ethylfuran	29	11	93	2	115	5
2,5-Dimethylfuran	46	11	98	2	120	4
2-Pentylfuran	-	-	83	3	83	11

*Accuracy was determined as follows: ((measured concentration – concentration in blank)/spiked concentration)*100

Conclusion

A HS-SPME-GC-MS method was developed for the analysis of furans and alkyl furans in baby formula and coffee. Different experimental conditions were evaluated and optimized. Satisfactory results in terms of linearity, accuracy and precision were obtained in the majority of the cases. Accuracy values above 111% in coffee samples spiked at 4000 µg/kg could be due to sample handling, but additional experimental work may be needed to further understand this bias.

- Pawliszyn, J., Handbook of Solid Phase Microextraction. Chemical Industry Press, Beijing 2009.
- Frank, N., Dubois, M., Huertas Pérez, J. F., Detection of Furan and five Alkylfurans, including 2-Pentylfuran, in various Food Matrices. J. Chromatogr. A 2020, 1622, 461119.