Forensic Toxicol (2015) 33:37–44
DOI 10.1007/s11419-014-0244-7

ORIGINAL ARTICLE

Hair analysis of synthetic cannabinoids: does the handling of herbal mixtures affect the analyst's hair concentration?

Bjoern Moosmann · Tsvetelina Valcheva · Merja A. Neukamm · Verena Angerer · Volker Auwärter

Received: 28 August 2014 / Accepted: 16 September 2014 / Published online: 7 October 2014

Abstract When narcotics police officers or other persons handling drug materials at work are suspected of consuming drugs, hair analysis may be useful to prove or refute such suspicion. However, it is known for many drugs that differentiation between actual drug use and external contamination can be challenging or sometimes impossible. This study evaluated the extent of external contamination caused by handling of synthetic cannabinoid-containing materials under realistic conditions in a forensic laboratory. Hair samples of laboratory staff were systematically analyzed for synthetic cannabinoids with a validated liquid chromatography–tandem mass spectrometry method after a large quantity of seized "legal high" products was analyzed in our laboratory. Furthermore, hair samples of laboratory staff not directly in contact with the drug materials and close relatives of exposed subjects were analyzed to check for cross contamination. All samples of persons who were in direct contact with drug materials tested positive for at least one synthetic cannabinoid. Concentrations ranged from trace amounts up to a maximum of 170 pg/mg (JWH-210), and roughly reflected the duration and intensity of exposure. Unexpectedly, subjects without direct contact with drug material also showed measurable concentrations of synthetic cannabinoids in hair. Concentrations caused by contamination were within the typical range found in known users of these drugs and could lead to false positive results and incorrect conclusions. Therefore, we recommend that body fluids should be simultaneously analyzed to unambiguously prove use of these drugs.

Keywords Synthetic cannabinoid · Legal highs · Hair analysis · External contamination · LC–MS–MS · Laboratory staff

Introduction

Synthetic cannabinoids, often referred to as "cannabinoid receptor agonists", are relatively new analytes in hair analysis with only a few methods published in the literature so far [1–4]. This category of drugs was first identified in 2008 in Germany and Japan [5, 6]; these novel psychoactive substances (NPS) mark the largest group of compounds reported through the Early Warning System of the European Monitoring Centre for Drugs and Drug Addiction (EMCCDA) in the past few years, with 30 new compounds in 2012 [7] and 29 in 2013 [8]. Because of the high market dynamics and frequent appearances of new compounds, it remains challenging to promptly include metabolites of the extensively metabolized synthetic cannabinoids into analytical methods [9]. Furthermore, it remains unclear whether the metabolites are incorporated into hair from the bloodstream to any significant degree. As a consequence, the parent compounds are the usual targets in hair analysis for synthetic cannabinoids. However, similar to other drugs of abuse that are smoked and handled extensively prior to consumption, external contamination may distort the

B. Moosmann · T. Valcheva · M. A. Neukamm · V. Angerer · V. Auwärter (✉)
Forensic Toxicology Department, Institute of Forensic Medicine, Medical Center - University of Freiburg, Albertstr 9, 79104 Freiburg, Germany
e-mail: volker.auwaerter@uniklinik-freiburg.de

B. Moosmann · V. Angerer
Hermann Staudinger Graduate School, University of Freiburg, Freiburg, Germany

results [10, 11]. In numerous cases with confirmed heavy consumption, high concentrations of parent compounds are found in the hair, but no metabolites could be detected (own unpublished data). However, a few authors have reported the detection of synthetic cannabinoid metabolites in the hair samples of alleged consumers [12, 13].

To distinguish between incorporation through the bloodstream and external contamination, Tsanaclis et al. [14, 15] suggested analyzing the washing solutions. If the ratio of a total analyte content in the wash solution to a total analyte content in the hair sample is less than 0.5, then pronounced external contamination is considered to be unlikely.

Apart from drug consumers, traffickers, and dealers, another group of people having contact with drug materials are narcotics police officers or analysts handling seized samples at work. If individuals of the latter groups are suspected of consuming drugs, hair analysis may be used to prove or refute such suspicion. Furthermore, because contamination of hair is not only possible through exposure to powder particles but also through contaminated fingers and hands, the question arises whether these issues extend to persons living in close contact with a person handling drugs at work. To evaluate the extent of external contamination caused by handling of synthetic cannabinoid containing drug materials under realistic conditions in a forensic laboratory, a study was performed on hair samples of people that were involved in the analysis of 670 herbal mixture samples within a 2-week period. In addition, hair samples of laboratory staff not directly in contact with the drug materials and hair samples of close relatives of exposed subjects were analyzed to check for cross contamination.

Materials and methods

Chemicals and reagents

Formic acid (Rotipuran® ≥98 %) was purchased from Carl Roth (Karlsruhe, Germany). Acetonitrile, ammonium formate (99.995 %), and ethanol (analytical grade) were from Sigma Aldrich (Steinheim, Germany). Deionized water was prepared using a cartridge deionizer from Memtech (Moorenweis, Germany). UR-144 and XLR-11 were kindly provided by Ilmari Szilvay (Finnish Customs Laboratory,

Table 1 Details of study participants including exposure status, time of hair sample collection and number of hair washes

Participant	Group[a]	Working hours	Time between last exposure and sampling (days)	Hair washes between last exposure and sampling	Hair length (cm)	Alternative samples
1	A	60	1	1	8	Leg hair
			14	12	7	
2	A	Coordinator[b]	2	2	0.5	Beard, chest, and leg hair
			8	8	0.5	
3	A	4	2	1	11.5	Pubic, leg, chest, and axillary hair
			10	8	10.5	
4	A	30	5	5	10.5	
5	A	30	3	2	39	
6	A	10	5	3	28	
7	A	3	10	10	5	
8	A	Coordinator[b]	2	1	16	
			5	3	17	
9	B	Household of participant 2	6	Unknown	34	
10–13	B	Household of participants 1 and 3	14	Unknown	3–27	
14	B	Household of articipants 1 and 3	14	Unknown	42	
15–23	C	Laboratory staff	5	Unknown	4.5–50	

[a] Group A: participants involved in analysis of the herbal mixtures; group B: participants living in the household of a group A participant; group C: laboratory members not involved in analysis of the herbal mixtures

[b] Two coordinators were frequently present in the room where the weighing out and extraction was performed and also carried out these tasks

Espoo, Finland). MAM-2201 ([1-(5-fluoropentyl)-1*H*-indol-3-yl](4-methyl-1-naphthalenyl)-methanone) was extracted from a herbal mixture [16, 17]. RCS-4 [(4-methoxyphenyl)(1-pentyl-1*H*-indol-3-yl)methanone] was purchased from Cayman Chemical (Ann Arbor, MI, USA). JWH-081 [(4-methoxy-1-naphthalenyl)(1-pentyl-1*H*-indol-3-yl)-methanone], JWH-122 [(4-methyl-1-naphthalenyl)(1-pentyl-1*H*-indol-3-yl)-methanone], JWH-210 [(4-ethyl-1-naphthalenyl)(1-pentyl-1*H*-indol-3-yl)-methanone], JWH-307 [5-(2-fluorophenic)-1-pentylpyrro(-3-yl)-naphthalene-1-yl-methanone], AM-2201 ([1-(5-fluoropentyl)-1*H*-indol-3-yl]-1-naphthalenyl-methanone), and AM-2232 [3-(1-naphthalenylcarbonyl)-1*H*-indole-1-pentanenitrile] were provided by the German Federal Criminal Police Office (BKA), the State Bureaus of Criminal Investigation (LKA) of Baden-Württemberg (Stuttgart, Germany) and Niedersachsen (Hannover, Germany), or purchased as "research chemicals" over the Internet. ^{1}H and ^{13}C nuclear magnetic resonance spectroscopy (NMR), gas chromatography–mass spectrometry (GC–MS), and thin-layer chromatography (TLC) were used to verify the identity and purity (≥98 %) of substances not obtained from professional vendors. The deuterated analogues JWH-018-d_{11} was obtained from Chiron AS (Trondheim, Norway) and JWH-081-d_9, JWH-120-d_9, JWH-210-d_9, MAM-2201-d_5, RCS-4-d_9, UR-144-d_5, and XLR-11-d_5 were purchased from Cayman Chemical for use as internal standards (ISs).

Sampling

Three groups of participants were included in the study. Group A consisted of eight participants who were involved in the analysis of herbal mixtures seized from an online retailer. Group B included five persons living in the same households with participants from group A. Group C comprised nine participants from laboratory staff not directly in contact with the drug materials. Head hair samples were collected from all participants from the posterior vertex region as close to the scalp as possible (remaining hair length at the scalp approximately 1 mm). For participants 1–3 and 8, scalp hair was sampled twice. Furthermore, hair samples from alternative sampling sites were obtained from three participants. The samples were stored in darkness at room temperature until analysis. The time periods between last exposure and the samplings are also given in Table 1 and ranged from 2 to 14 days.

Handling and sample preparation of herbal mixtures carried out by participants (group A)

The herbal mixtures analyzed by the participants were part of a seizure that included more than 4,000 samples from an online vendor selling "legal highs". Six hundred and seventy samples covering 31 brands were weighed, extracted, and filtered prior to analysis by the participants as described elsewhere [18]. Gloves and laboratory coats were worn during the entire sample preparation period. Furthermore, weighing and extraction were carried out in a dedicated room, separated from sample preparation of human specimens. The respective herbal mixtures contained 12 different synthetic cannabinoids (AM-1220, AM-2201, AM-2232, JWH-081, JWH-122, JWH-203, JWH-210, JWH-307, MAM-2201, RCS-4, UR-144, XLR-11) at concentrations ranging from 1.8 to 18.9 % (w/w), and were analyzed within 2 weeks.

Hair sample preparation

Depending on hair length and the amount of sample available, the hair strands were either segmented or analyzed as a whole (hair lengths and segmentation are given in Tables 1 and 3). Prior to extraction, the hair was washed by shaking for 4 min with 4 ml of water, followed by 4 ml of acetone, and 4 ml of petroleum ether, respectively. After allowing the hair to dry for 24 h, 50 mg of the sample was cut into pieces of 1–2 mm.

Table 2 Multiple reaction monitoring transitions and corresponding voltages applied for liquid chromatography–tandem mass spectrometry analysis

Analyte	Q1	Q3	DP (V)	CE (V)	CXP (V)	IS
AM-2201	360	155	85	36	11	JWH-018-d_{11}
		127		64	8	
AM-2232	353	155	80	33	12	JWH-018-d_{11}
		127		70	9	
JWH-081	372	185	70	37	13	JWH-081-d_9
		214		61	10	
JWH-122	356	169	94	36	12	JWH-122-d_9
		141		66	9	
JWH-210	370	183	80	35	8	JWH-210-d_9
		214		35	10	
JWH-307	386	155	60	30	11	JWH-081-d_9
		127		72	9	
MAM-2201	374	169	70	37	13	MAM-2201-d_5
		141		61	10	
RCS-4	322	135	80	34	10	RCS-4-d_9
		77		77	4	
UR-144	312	125	80	34	16	UR-144-d_5
		214		34	17	
XLR-11	330	125	85	32	9	XLR-11-d_5
		232		34	18	

Q1 *m/z* of the precursor ion, *Q3* *m/z* of the product ion, *DP* declustering potential, *CE* collision energy, *CXP* collision cell exit potential, *IS* internal standard

Table 3 Synthetic cannabinoid concentrations detected in hair segments obtained from group A participants (participants 1–8) and group B participants (participants 9 and 14)

Participant	Segment (cm)	Time between last exposure and sampling (days)	JWH-081 (pg/mg)	JWH-122 (pg/mg)	JWH-210 (pg/mg)	JWH-307 (pg/mg)	AM-2201 (pg/mg)	AM-2232 (pg/mg)	MAM-2201 (pg/mg)	RCS-4 (pg/mg)	UR-144 (pg/mg)	XLR-11 (pg/mg)
1	0–3	1		0.8	25	3.1						0.9
	3–8		<0.5	7.9	170	43	0.5		1.6			6.5
	0–3	14		<0.5	4.5	0.8						
	3–7			6.0	160	25	<0.5		<0.5			
2	Full length	2			<0.5							
	Full length	8										
3	0–6	2		1.0	33	2.9	0.9					
	6–11.5		<0.5	2.0	66	8.3	0.8					
	0–3	10		<0.5	11	1.4	<0.5					
	3–6			1.2	38	5.5	<0.5					
	6–10.5			3.0	71	6.8	0.6					
4	0–6	5	5.5	5.1	3.1	1.4	0.7	1.5	24	<0.5		2.6
	6–10.5		0.7	21	12	2.5	3.0	1.7	27		0.7	
5	0–3	3	<0.5	<0.5	1.0	1.6				<0.5		
	3–6		<0.5	1.1	4.1	6.3		<0.5	1.1	0.6		
	6–9			0.6	1.8		2.0	<0.5		<0.5		
	9–12		<0.5	0.9		3.0	0.5	<0.5	0.7	<0.5		
	12–18		<0.5	1.1	2.1	2.1	0.7	<0.5	0.9	1.0		
	18–39		<0.5	2.5	6.6	3.5	1.4	0.8	0.7	0.9		
6	0–6	5			1.0							
	6–28											
7	Full length	10			<0.5	<0.5	<0.5					
8	0–3	2		1.0		<0.5						
	3–6			6.3	4.8	1.1	<0.5	0.5				
	6–16			8.7	6.6	1.5	1.5	1.2				
	0–3	5		0.9	<0.5	0.6						
	3–6			4.6	5.3	4.2						
	6–17			13	9.1	6.9	3.2		0.6			
9	0–6	6										
	6–12				7.9							
	12–34				11							
14	0–6	14			<0.5							
	6–12				<0.5							
	12–18											
	18–42											

Entries with "< 0.5" were below the lower limit of quantitation (LLOQ); no entry indicates that the analyte was not detected

For extraction, 1.5 ml of ethanol as well as 20 µl of IS solution were added. The analytes were then extracted for 3 h under sonication. Finally, 1 ml of the extract was transferred into a vial, evaporated to dryness, and reconstituted in 100 µl of mobile phase A/B (50:50, v/v) prior to liquid chromatography–tandem mass spectrometry (LC–MS–MS) analysis [2].

LC–MS–MS method

A fully validated LC–MS–MS method, covering all synthetic cannabinoids detected in the investigated herbal mixtures, was applied. The lower limit of quantitation (LLOQ) for the respective analytes was 0.5 pg/mg. The

instrument consisted of a Shimadzu Prominence HPLC system (Shimadzu, Duisburg, Germany) coupled to a QTrap 4000 tandem mass spectrometer (AB Sciex, Darmstadt, Germany). Chromatographic conditions were as published by Huppertz et al. [19]. In brief, separation was performed on a Kinetex C18 column (100 × 2.1 mm i.d., particle size 2.6 µm) with a corresponding guard column (Phenomenex, Aschaffenburg, Germany). Solvent A consisted of 0.1 % formic acid, 2.0 mM ammonium formate, and 1 % acetonitrile in water; solvent B consisted of 0.1 % formic acid and 2.0 mM ammonium formate in acetonitrile. Gradient elution was applied starting at 20 % mobile phase B for 1 min, increased to 60 % B within 1.5 min, further increased to 65 % B within 1.5 min, held for 1.5 min, increased to 99 % B within 2.5 min, and held for 2 min. Starting conditions were restored within 0.2 min, and the system was re-equilibrated for 1.8 min prior to the injection of the next sample. The flow rate was set to 0.5 ml/min; the injection volume was 20 µl, and the auto-sampler and column oven temperatures were 10 and 40 °C, respectively. The mass spectrometer was operated in positive electrospray ionization (ESI) mode. The ion source voltage was set to 2,500 V and the ion source temperature was 600 °C. The gas pressures were: curtain gas (N_2), 207 kPa; ion source gas 1 and 2 (compressed air), 276 and 345 kPa, respectively; collision gas (N_2), 41 kPa.

The MS was operated in scheduled multiple reaction monitoring (sMRM) mode, including the transitions listed in Table 2.

Analysis of wash solutions

A 2-ml volume of each acetone or petroleum ether wash solution was analyzed by LC–MS–MS as described above utilizing three-point calibration curves (1, 2.5, and 75 ng/ml) in acetone or petroleum ether wash solution with drug-free hair.

Results and discussion

Synthetic cannabinoid concentrations measured for segments or full lengths of hair obtained from people of different groups

All samples from persons who were in direct contact with drug materials (group A) tested positive for at least one synthetic cannabinoid (JWH-081, JWH-122, JWH-210, JWH-307, AM-2201, AM-2232, MAM-2201, XLR-11, RCS-4, or UR-144). Concentrations ranged from trace amounts up to a maximum of 170 pg/mg (JWH-210). Exact values are given in Table 3. In addition, one of the samples obtained from laboratory staff not involved in the analysis was positive (0.9 pg/mg for JWH-210). This could be due to cross contamination originating from the herbal mixture analysis. Comparing the concentrations detected in the hair samples of five participants with different work durations, the concentrations roughly reflected the duration of exposure.

Unexpectedly, subjects without direct contact with drug materials also showed measurable concentrations of synthetic cannabinoids in hair. One household group (participants 10–14) included two putative donors of contamination, viz., the husband/father (participant 3) and one child/sibling (participant 1). In this household, one child/sibling (participant 14) had trace amounts (<0.5 pg/mg) of JWH-210 in the two proximal 6-cm-long segments (42 cm hair strand), and no synthetic cannabinoids were detected in the hair of the wife/mother or the three other children/siblings. While these trace amounts do not raise serious concerns, the results for another household (participant 9) were very surprising. Despite a JWH-210 concentration of less than 0.5 pg/mg in the hair sample of participant 2, who was involved in the work, up to 11 pg/mg was detected in the hair sample of his girlfriend (participant 9), who lived in the same household, but had no contact with the drug materials (Table 3). One possible explanation for these results could be direct transfer through contaminated fingers; for example, from a head massage or by sleeping on pillows accidentally contaminated by the hands of the partner. The presence of only trace amounts of cannbinoids in the hair samples of participant 2 was attributed to his short hair length (0.5 cm), because previous studies have indicated that shorter hair is less prone to external contamination [10]. Obviously, the risk of cross contamination would be much higher if the drugs were handled at home.

Table 4 Synthetic cannabinoid concentrations in hair samples obtained from alternative sampling sites

Participant	Sampling site	Time between last exposure and sampling (days)	JWH-122 (pg/mg)	JWH-210 (pg/mg)	JWH-307 (pg/mg)
1	Left leg	1	0.5	2.8	2.2
	Right leg		<0.5	1.7	2.1
2	Beard	2	<0.5	4.7	1.3
	Chest				
	Upper leg				
3	Left leg	2			
	Right leg				
	Armpit				
	Chest			<0.5	
	Pubic region		0.8	22	1.6

Entries with "<0.5" were below LLOQ; no entry indicates that the analyte was not detected

Table 5 Ratios of the total amount of synthetic cannabinoid in each acetone wash solution to that in each hair sample of participants 1–9

Participant	Segment (cm)	Time between last exposure and sampling (days)	JWH-081	JWH-122	JWH-210	JWH-307	AM-2201	AM-2232	MAM-2201	RCS-4	UR-144	XLR-11
1	0–3	1		23	9.9	11						0
	3–8		0	4.6	3.0	1.8	0		0			0
	0–3	14		0	0	0						
	3–7			0.3	0.2	0	0		0			
2	Full length	2			Pos							
	Full length	8										
3	0–6	2		4.6	3.2	4.0	0					
	6–11.5		0	2.4	1.5	2.4	0					
	0–3	10		0	0.6	0	0					
	3–6			0.5	0.6	0.8	0					
	6–10.5			0.5	0.4	0.5	0					
4	0–6	5	0	0	0	0	0	0	0	0		0
	6–10.5		0	3.0	2.5	2.8	0	5.6	2.9		0	
5	0–3	3	Pos	Pos	14	11				0		
	3–6		0	16	9.4	12		0	11	0		
	6–9			18	13		0	0		0		
	9–12		0	9.0		15	0	0	7.3	0		
	12–18		Pos	17	18	24	0	0	12	0		
	18–39		0	0	0	0	0	0	0	0		
6	0–6	5			0							
	6–28											
7	Full length	10			0	0	0					
8	0–3	2		30		Pos						
	3–6			4.0	1.9	0	0	0				
	6–16			8.5	3.8	8.4	0	0				
	0–3	5		2.2	3.6	0						
	3–6			0	0	0						
	6–17			1.8	0	2.7	0		0			
9	0–6	6										
	6–12				0							
	12–34				0							

Entries "0" represent negative findings in the wash solution but positive findings in the hair extract. No entry indicates negative findings in the wash solution and the hair sample for the respective analyte

Pos positive result with concentration of analyte in hair below LLOQ (0.5 pg/mg)

Washout effects

Four participants from group A (participants 1–3 and 8) provided two hair samples from different time points after exposure. The time period between first and second sampling ranged from 3 to 13 days. In general, synthetic cannabinoid concentrations were similar for the two sampling time points (Table 3). However, conclusions based on this observation should be treated with caution because it is likely that an external contamination would not be distributed evenly over the head [10].

Alternative sampling sites

Analysis of hair samples obtained from other areas of the body showed that the issue of external contamination also applies to body parts that were covered by clothing during the handling of drug material (Table 4). For participant 1, three synthetic cannabinoids were detected at low concentrations in the leg hair samples taken from both legs. Three synthetic cannabinoids were detected in the beard hair of participant 2, who had only trace amounts of JWH-210 in his scalp hair samples. Chest and upper-leg hair

Table 6 Ratios of the total amount of synthetic cannabinoid in each petroleum ether wash solution to that in each hair sample of participants 1–9

Participant	Segment (cm)	Time between last exposure and sampling (days)	JWH-081	JWH-122	JWH-210	JWH-307	AM-2201	AM-2232	MAM-2201	RCS-4	UR-144	XLR-11
1	0–3	1		14	4.5	7.4						0
	3–8		0	3.2	3.0	0	0		1.5			0
	0–3	14		0	0	0						
	3–7			0	0	0	0		0			
2	Full length	2			0							
	Full length	8										
3	0–6	2		6.2	0.7	3.1	0					
	6–11.5		0	5.5	0.4	1.9	0					
	0–3	10		0	0.1	0						
	3–6			0	0.1	0	0					
	6–10.5			0	0	0	0					
4	0–6	5	0	0.2	0	0	0	0	0	0		0
	6–10.5		0	0.5	0.3	0	0	0	0			
5	0–3	3	0	16	0	6.4				0		
	3–6		0	8.8	2.8	4.1		0	3.4	0		
	6–9			14	3.6		0	0		0		
	9–12		0	7.6		4.7	0	0	0	0		
	12–18		0	9.3	0	10	0	14	0	0		
	18–39		0	5.4	3.1	4.5	0	18	0	0		
6	0–6	5			0							
	6–28											
7	Full length	10			Pos	0	0					
8	0–3	2		8.0		Pos						
	3–6			1.8	0.8	6.3	0					
	6–16			1.6	0.7	4.4	0					
	0–3	5		0	0	0						
	3–6			0	0	0						
	6–17			0	0	0.2	0		0			
9	0–6	6										
	6–12					0						
	12–34					0						

Entries "0" represent negative findings in the wash solution but positive findings in the hair extract. No entry indicates negative findings in the wash solution and the hair sample for the respective analyte

Pos positive result with concentration of analyte in hair below LLOQ (0.5 pg/mg)

samples were negative for this participant. The higher concentrations in beard hair, which was similar in length to the scalp hair, may be a result of less efficient incorporation of the compound in scalp hair than in beard hair. A more likely explanation would be the transfer of compounds through contaminated fingers, because the participant reported touching his beard hair more frequently than his scalp hair. Participant 3 showed the highest concentration in pubic hair (22 pg/mg JWH-210) and trace amounts in his chest hair.

Wash solutions

The ratios of total analyte content in each wash solution to total analyte content in the corresponding hair sample ranged from 0 to 48 in the acetone solutions (Table 5), and from 0 to 18 in petroleum ether solutions (Table 6). The high ratios for some of the samples strongly suggest external contamination. However, not all samples had such high ratios and in some cases, for example, participant 9, who was not involved in the work, no analyte was detected

in the wash solutions. Comparing the ratios in the participants that provided two samples at different time points (participants 1–3 and 8), the ratios declined drastically for all four participants, and the ratio was below 0.1 in the second sample for most analytes and segments. These results suggest that the drugs become incorporated into the hair matrix over time by diffusion across the cell membrane complex and are not removed by wash procedures.

Conclusions

Depending on the duration and intensity of exposure, considerable concentrations of synthetic cannabinoids were found in hair samples of persons exposed to these drugs at work. Unexpectedly, cross contamination from an exposed person to a close relative occurred and led to (false) positive results. The concentrations caused by the contamination are in the typical range found in known users of these drugs and could lead to incorrect conclusions in forensic cases or workplace drug testing. Therefore, the analysis of body fluids is strongly advised to unambiguously prove the use of these drugs, particularly when metabolites are not detected in hair samples.

Acknowledgments The authors thank the Drug Prevention and Information Programme of the European Union (JUST/2011/DPIP/AG/3597), the German Federal Ministry of Health, and the City of Frankfurt/Main for financial support.

Conflict of interest There are no financial or other relations that could lead to a conflict of interest.

References

1. Gottardo R, Sorio D, Musile G, Trapani E, Seri C, Serpelloni G, Tagliaro F (2014) Screening for synthetic cannabinoids in hair by using LC-QTOF MS: a new and powerful approach to study the penetration of these new psychoactive substances in the population. Med Sci Law 54:22–27
2. Hutter M, Kneisel S, Auwärter V, Neukamm MA (2012) Determination of 22 synthetic cannabinoids in human hair by liquid chromatography-tandem mass spectrometry. J Chromatogr B 903:95–101
3. Salomone A, Gerace E, D'Urso F, Di Corcia D, Vincenti M (2012) Simultaneous analysis of several synthetic cannabinoids, THC, CBD and CBN, in hair by ultra-high performance liquid chromatography tandem mass spectrometry. Method validation and application to real samples. J Mass Spectrom 47:604–610
4. Salomone A, Luciano C, Di Corcia D, Gerace E, Vincenti M (2014) Hair analysis as a tool to evaluate the prevalence of synthetic cannabinoids in different populations of drug consumers. Drug Test Anal 6:126–134
5. Auwärter V, Dresen S, Weinmann W, Müller M, Pütz M, Ferreiros N (2009) 'Spice' and other herbal blends: harmless incense or cannabinoid designer drugs? J Mass Spectrom 44:832–837
6. Uchiyama N, Kikura-Hanajiri R, Kawahara N, Haishima Y, Goda Y (2009) Identification of a cannabinoid analog as a new type of designer drug in a herbal product. Chem Pharm Bull 57:439–441
7. European Monitoring Centre for Drugs and Drug Addiction (2012) The state of the drugs problem in Europe, annual report 2012. Publications Office of the European Union, Lisbon
8. European Monitoring Centre for Drugs and Drug Addiction (2014) European drug report 2014: trends and developments. Publications Office of the European Union, Lisbon
9. De Brabanter N, Espostio S, Geldof L, Lootens L, Meuleman P, Leroux-Roels G, Deventer K, Van Eenoo P (2013) In vitro and in vivo metabolism of 1-pentyl-3-(4-methyl-1-nephthoyl)indole (JWH-122). Forensic Toxicol 31:212–222
10. Moosmann B, Roth N, Auwärter V (2014) Hair analysis for THCA-A, THC and CBN after passive in vivo exposure to marijuana smoke. Drug Test Anal 6:119–125
11. Pragst F, Balikova MA (2006) State of the art in hair analysis for detection of drug and alcohol abuse. Clin Chim Acta 370:17–49
12. Kim J, In S, Park Y, Park M, Kim E, Lee S (2013) Deposition of JWH-018, JWH-073 and their metabolites in hair and effect of hair pigmentation. Anal Bioanal Chem 405:9769–9778
13. Salomone A, Gerace E, Luciano C, Di Corcia D, Vincenti M (2014) Quantification of 22 synthetic cannabinoids and 10 metabolites in human hair. Toxicol Anal Clin 26:S14
14. Tsanaclis L, Nutt J, Bagley K, Bevan S, Wicks J (2014) Differentiation between consumption and external contamination when testing for cocaine and cannabis in hair samples. Drug Test Anal 6:37–41
15. Tsanaclis L, Wicks JF (2008) Differentiation between drug use and environmental contamination when testing for drugs in hair. Forensic Sci Int 176:19–22
16. Moosmann B, Kneisel S, Girreser U, Brecht V, Westphal F, Auwärter V (2012) Separation and structural characterization of the synthetic cannabinoids JWH-412 and 1-[(5-fluoropentyl)-1H-indol-3yl]-(4-methylnaphthalen-1-yl)methanone using GC-MS, NMR analysis and a flash chromatography system. Forensic Sci Int 220:e17–e22
17. Moosmann B, Kneisel S, Wohlfarth A, Brecht V, Auwärter V (2013) A fast and inexpensive procedure for the isolation of synthetic cannabinoids from 'Spice' products using a flash chromatography system. Anal Bioanal Chem 405:3929–3935
18. United Nations Office on Drugs and Crime (2013) Recommended methods for the identification and analysis of synthetic cannabinoid agonists in seized materials. UNODC, Vienna
19. Huppertz LM, Kneisel S, Auwärter V, Kempf J (2014) A comprehensive library-based, automated screening procedure for 46 synthetic cannabinoids in serum employing liquid chromatography-quadrupole ion trap mass spectrometry with high-temperature electrospray ionization. J Mass Spectrom 49:117–127