

Establishment of analytical methods for allergenic compounds in mouthwashes and sanitary napkins by ultra-high-performance liquid chromatography with tandem mass spectrometry

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Abstract: Analytical methods for detecting atranol, chloroatranol, evernic acid, (+)-usnic acid, and atranorin in sanitary napkins and mouthwashes were developed using ultrahigh performance liquid chromatography-tandem mass spectrometry (UHPLC-MS/MS). UHPLC-MS/MS conditions were optimized for rapid, sensitive, and simultaneous analysis of the five allergenic compounds. The methods were validated by assessing their specificity, matrix effects, limit of detection (LOD), limit of quantification (LOQ), linearity, accuracy, and precision. Good linearity was achieved with a determination coefficient of ≥ 0.99 . The LOD and LOQ were 2.1-9.8 and 6.4-29.6 ng/g for sanitary napkins and 0.29-0.48 and 0.87-1.45 ng/mL for mouthwashes, respectively. The accuracy and precision were within an acceptable range according to the criteria reported in the European SANTE/11813/2017 guidelines (70-120 % recovery, < 20 % relative standard deviation). Therefore, these methods can be used to analyze atranol, chloroatranol, evernic acid, (+)-usnic acid, and atranorin in sanitary napkins and mouthwashes.

Key words: allergenic compounds #1, mouthwash #2, sanitary napkins #3, tandem mass spectrometry #4, method validation #5

1. Introduction

Mouthwash is an auxiliary oral hygiene product used to clean the oral cavity by decreasing the number of microorganisms and removing bad breath and plaque. According to recent research, povidone-iodine mouthwashes can reduce the risk of cross-infection of coronavirus disease (COVID-19),¹⁻³ making them

an essential personal hygiene product in the post-COVID-19 era. Sanitary napkins are used cyclically by women of childbearing age during menstruation, and constitute an important category of feminine hygiene products.⁴ However, serious concerns were raised in Korea regarding the presence of volatile organic compounds in sanitary napkins and humidifier disinfectants, which spread chemophobia among

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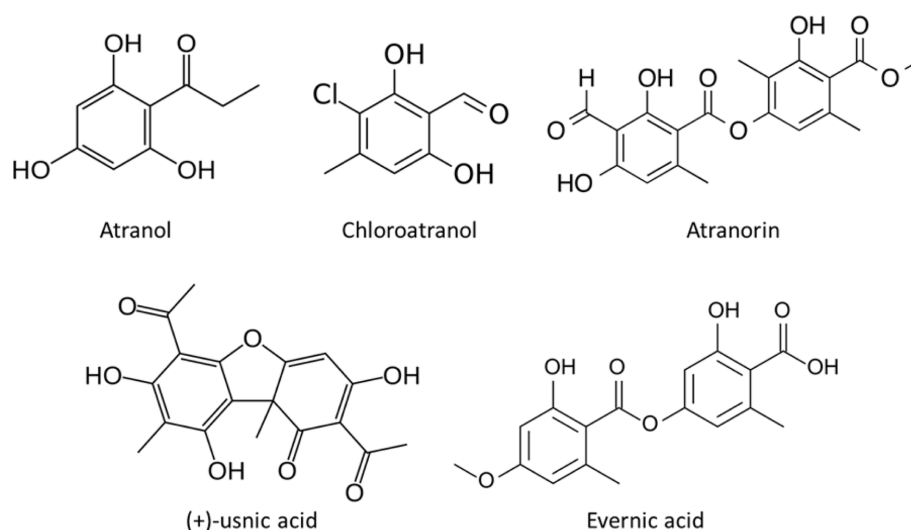


Fig. 1. Chemical structures of five allergenic compounds.

consumers. The distrust of consumers stems from their anxiety, which arises from the absence of information about raw materials and additives in personal hygiene products. Therefore, the safety and necessity for the indication of all ingredients of commercially available personal hygiene products are becoming critical societal issues.

Numerous cases of allergic contact dermatitis caused by using sanitary napkins and mouthwashes have been reported as these personal hygiene products are directly in contact with the skin.⁵⁻¹⁰ The known reasons of contact dermatitis are the antifungal agents, preservatives, colophony, adhesives, and fragrances.^{6,7,11} Several studies have reported the presence of fragrances in absorbent hygiene products, including sanitary napkins, tampons, and panty liners,¹² and oral hygiene products, including toothpastes, mouthwashes, and dental flosses.¹³ However, the presence of these fragrances is not communicated to customers.¹²

The European Union (EU) regulates tolerance limits for allergenic fragrance ingredients, and has suggested guidelines for cosmetic product labeling and announced a list of substances that cosmetic products must not contain in Annex III of EC regulation No. 1223/2009.¹⁴ In 2019, the Ministry of Food and Drug Safety (MFDS) published an administrative notice of regulations regarding indications of quasi-drugs: the

names of the allergenic compounds should be specified when the quasi-drug contains allergenic compounds included in the list announced by the MFDS.

Five chemicals derived from two natural extracts (oak moss and tree moss extracts): atranol, chloroatranol, atranorin, evernic acid, and usnic acid, which were listed by the EU and MFDS as mentioned above, were selected as the analytes in this study. The chemical structures of the analytes are shown in Fig. 1. The analytes are known as allergens.¹⁵⁻¹⁷ Atranol and chloroatranol are representative sensitizers present in the two natural extracts, and their use in cosmetic products is prohibited by the EU Commission.¹⁴ The analytical method for quantifying these two sensitizers was described by the International Fragrance Association.¹⁸ In addition, several analytical methods using liquid chromatography-mass spectrometry (LC-MS)¹⁹⁻²³ and gas chromatography-mass spectrometry (GC-MS)^{24,25} have been reported. Musharraf *et al.*¹⁹ identified and quantified several secondary metabolites, including atranorin and usnic acid, in lichen extracts using LC coupled with triple-quadrupole tandem MS. Goursot²⁰ suggested two simultaneous analytical methods for atranol and chloroatranol in moss extracts using LC with photodiode array detection and LC-MS for quality control laboratories. Several studies have determined atranol

and chloroatranol in perfumes using LC-MS/MS.^{21,22} López-Nogueroles *et al.*²⁵ developed a new GC-MS method for quantifying atranol and chloroatranol in perfumes based on simultaneous derivatization and dispersive liquid-liquid microextraction to remove and concentrate polar compounds. Additionally, several qualitative analysis methods have been reported for compounds present in oakmoss absolute²³ and lichen.²⁴ The majority of studies have analyzed numerous allergenic compounds in moss (lichen) extracts and cosmetic products. However, to the best of our knowledge, no study has simultaneously identified the five aforementioned allergenic compounds in personal hygiene products.

In this study, we developed a method for the simultaneous quantification of the five sensitizers in sanitary napkins and mouthwashes. The sonication and liquid-liquid extraction methods were conducted for sanitary napkins and mouthwash samples with several salts, respectively, to extract the five allergenic compounds from each matrix, and optimized ultrahigh performance liquid chromatography-tandem mass spectrometry (UHPLC-MS/MS) conditions were used to analyze the five sensitizers in two different matrices. The method described in this study is rapid (10 min, including column equilibration time), highly sensitive (ng/g, analyte/matrix), and validated with respect to specificity, linearity, matrix effect, accuracy, and precision.

2. Experimental

2.1. Chemicals and reagents

Atranol and chloroatranol standards were purchased from Sigma-Aldrich (St. Louis, MO, USA). Vanillin-(phenyl-¹³C₆), used as an internal standard (IS), was purchased from Sigma-Aldrich. The standards for evermic acid, (+)-usnic acid, and atranorin were obtained from Santa Cruz Biotechnology (Santa Cruz, CA, USA), PhytoLab (Vestenbergsgreuth, Bavaria, Germany), and BOC Sciences (Shirley, NY, USA), respectively. Acetic acid, ammonium formate, ammonium acetate, and formic acid were purchased from Sigma-Aldrich and purities were 99.7 %, 99.995 %,

99.0, and 98 %, respectively. HPLC-grade methanol, chloroform, water, and acetonitrile (ACN) were obtained from Honeywell Burdick & Jackson (Muskegon, MI, USA).

2.2. Preparation of standards solutions

Stock solutions of atranol, chloroatranol, and vanillin-(phenyl-¹³C₆) were prepared by dissolving pure standards in methanol. Chloroform was used to prepare the stock solutions of usnic acid and atranorin. A stock solution of evermic acid was prepared using methanol:chloroform (1:1, v/v) as the solvent. The stock solutions were stored at -20 °C until use.

2.3. Preparation of samples

Commercially available sanitary napkins and mouthwashes were purchased from local and online markets. The sanitary napkins were cut using scissors in a tray containing dry ice, and finely ground using a cryogenic mill (6870 Freezer/Mill, SPEX SamplePrep, Metuchen, NJ, USA) filled with liquid nitrogen. The cryogenically ground samples were maintained at -20 °C until extraction. Mouthwash samples were stored at room temperature (1-35 °C) until further extraction.

2.4. Pretreatment of samples

To analyze the five allergenic compounds in sanitary napkins, cryogenically ground samples (100 mg) were spiked with IS (20 µL; 5 µg/mL) and extracted by sonication with an extracting solvent (1 mL; methanol:chloroform, 1:1, v/v) for 15 min. The extracts were filtered through a polytetrafluoroethylene polymer (PTFE) syringe filter, whereafter the filtered extracts (0.5 mL) were dried using nitrogen gas. The dried samples were reconstituted using methanol:water (0.5 mL; 3:1, v/v) as the solvent, filtered through the PTFE syringe filter, and injected into the UHPLC-MS/MS system.

Mouthwash samples (0.5 mL) were spiked with IS (20 µL; 5 µg/mL), 0.4 M sodium chloride (0.5 mL), and dichloromethane (1 mL), and shaken three times for 30 s each. Following centrifugation for 3 min at 3000 rpm, the dichloromethane layers (0.8 mL) were

Table 1. Optimized reaction monitoring conditions for the five allergenic compounds and internal standard (IS)

Analytes (Abbreviation)	Monoisotopic Mass	Ionization type	DP	EP	Q1 (m/z)	Q3 (m/z)	CE
Atranol (Al)	152.0473	[M-H] ⁻	-60	-10	150.9	122.9 81.0	-20 -25
Chloroatranol (ChAl)	186.0084	[M-H] ⁻	-60	-10	184.9	156.7 92.9	-20 -28
(+)-Usnic acid (UA)	344.0896	[M-H] ⁻	-65	-10	343.0	328.0 259.0	-28 -28
Evernic acid (EA)	332.0896	[M-H] ⁻	-65	-10	330.9	166.7 149.1	-20 -30
Atranorin (An)	374.1002	[M-H] ⁻	-35	-10	373.2	176.8 162.8	-20 -29
Vanillin-(phenyl- ¹³ C ₆) (IS)	158.0675	[M-H] ⁻	-70	-10	156.9	142.0 96.9	-18 -28

transferred into new tubes, dried with nitrogen gas, and reconstituted using methanol:water (0.8 mL; 3:1, v/v) as the solvent. Prior to analysis, the extracts were filtered through the PTFE syringe filter.

2.5. UHPLC-MS/MS analysis

The UHPLC-MS/MS analysis was performed using a Nexera X2 UHPLC/HPLC system (Shimadzu, Japan) connected to a QTRAP 4500 mass spectrometer (SCIEX, USA). Chromatographic separation was performed using an Eclipse Plus C18 column (2.1 × 100 mm, 1.8 μm, Agilent, USA) connected to a C18 SecurityGuard ULTRA Cartridge (2.1 mm I. D., Phenomenex, USA). The flow rate, column temperature, and injection volume were set at 0.4 mL/min, 25 °C, and 2 μL, respectively. The four mobile phase compositions were tested as follows: 0.1 % acetic acid in water and 0.1 % acetic acid in ACN (MP1), 10 mM ammonium acetate containing 0.1 % acetic acid in water and 0.1 % acetic acid in ACN (MP2), 0.1 % formic acid in water and 0.1 % formic acid in ACN (MP3), and 10 mM ammonium formate containing 0.1 % formic acid in water and 0.1 % formic acid in ACN (MP4). The linear gradient elution was programmed as follows: 0-5 min, 25-95 % B; 5-7 min, isocratic 95 % B; 7-7.5 min, 95-25 % B; 7.5-10 min, isocratic 25 % B for column equilibration. The MS analysis was conducted in the negative ion mode with electrospray ionization. The

MS conditions were optimized as follows: ion spray voltage, -4500V; curtain gas, 30 psi; source temperature, 600 °C; ion source gas 1 and 2, 60 and 50 psi, respectively. In the selected reaction monitoring (SRM) mode, SRM transitions, declustering potential (DP), entrance potential (EP), and collision energy (CE) values for the five allergenic compounds and IS are listed in Table 1.

2.6. Method validation for sanitary napkin and mouthwash samples

Constructing a matrix-matched calibration curve and validating the method for quantifying the five allergenic compounds in sanitary napkins and mouthwash samples requires a matrix that does not contain the five analytes. To select a sanitary napkin for method validation, commercially available unscented sanitary napkins were preliminarily screened to investigate whether the five analytes were present in representative samples. The sanitary napkin that did not contain the five analytes was selected for method validation. In the case of mouthwash, four main ingredients of commercially available mouthwash samples, including 22.5 % ethanol, 10 % glycerin, 4 % xylitol, and 4 % hydrogenated castor oil, were mixed to prepare the matrix for matrix-matched calibration and method validation.

To evaluate the ME and construct a matrix-matched calibration curve, each matrix was extracted and

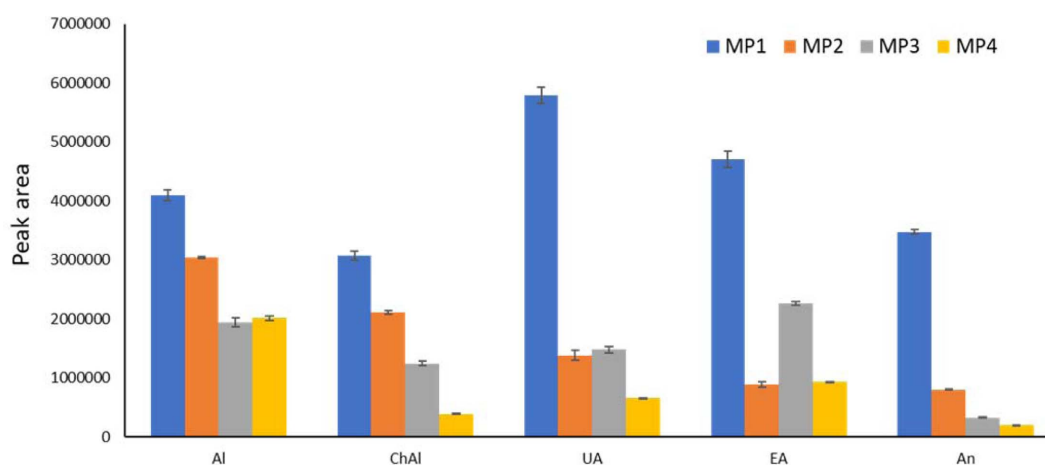


Fig. 2. Investigation of mobile phase compositions for the sensitive quantification of the five allergenic compounds by ultrahigh performance liquid chromatography–tandem mass spectrometry. Mobile phase (MP) compositions tested were as follows: MP1, 0.1 % acetic acid in water and 0.1 % acetic acid in acetonitrile (ACN); MP2, 10 mM ammonium acetate containing 0.1 % acetic acid in water and 0.1 % acetic acid in ACN; MP3, 0.1 % formic acid in water and 0.1% formic acid in ACN and; MP4, 10 mM ammonium formate containing 0.1 % formic acid in water and 0.1 % formic acid in ACN.

dried using the procedures mentioned in the section on the pretreatment of samples and reconstituted using solvent with the five analytes. LODs and LOQs were calculated based on the standard deviation of the response and the slope of the calibration curve.^{26,27} The formula for the LODs and LOQs used in this study were $LOD=3.3 \times \sigma/S$ and $LOQ=10 \times \sigma/S$, respectively, where σ is the standard deviation of y-intercepts of regression lines and S is the slope of the calibration curve. The accuracy and precision of the as-developed method were tested by spiking three different concentrations of the five analytes into the matrix prior to extraction. After spiking with standard solutions, the extraction and pretreatment steps mentioned in the section on sample pretreatment were conducted.

3. Results and Discussion

3.1. Optimization of UHPLC-MS/MS conditions for the analysis of five allergenic compounds

In this study, the composition of the mobile phase was optimized to simultaneously analyze the five allergenic compounds. Ammonium acetate, ammonium formate, acetic acid, and formic acid are commonly recommended additives for LC-MS analysis. In the

majority of cases, except for atranol in MP3 and MP4, the five analytes exhibited higher sensitivities in mobile phases lacking ammonium ions than in those containing ammonium ions. Upon comparing MP1 and MP3, all compounds demonstrated higher sensitivity in mobile phases containing acetic acid than those containing formic acid (Fig. 2). Based on these results, 0.1 % acetic acid in water and 0.1 % acetic acid in ACN were selected as the optimized mobile phase A and B, respectively.

The total time required for analysis of previously reported analytical methods^{20-22,24} for the quantification of allergenic compounds exceeded 30 min, except for a single study¹⁹ wherein the total run time was 6 min for the analysis of secondary metabolites of lichen, including two allergenic compounds (atranorin and (+) usnic acid) derived from the natural extracts announced in the EU regulation (No. 1223/2009). In this study, six peaks of analytes were detected within 6 min (Fig. 3). In addition, the total run time, including column equilibration time, for quantifying the five analytes and IS was 10 min, indicating that the methods described in this study are suitable for analyzing more sanitary napkin and mouthwash samples per unit time.

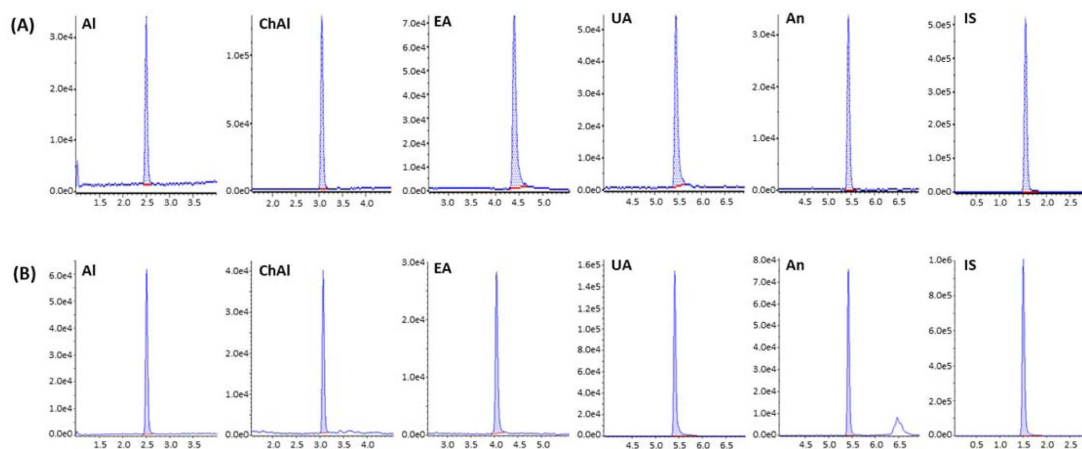


Fig. 3. Typical selected-reaction-monitoring chromatograms of the five allergenic compounds and internal standard (IS) spiked into the (A) sanitary napkin and (B) mouthwash extracts.

Table 2. Results of matrix effect for the five allergenic compounds in the unscented sanitary napkin and mouthwash matrix

	Sanitary napkin (% , n=3)						Mouthwash (% , n=3)					
	Low		Medium		High		Low		Medium		High	
	ME	RSD	ME	RSD	ME	RSD	ME	RSD	ME	RSD	ME	RSD
AI	93.5	1.2	102.2	0.9	106.2	1.2	79.0	7.4	75.6	11.8	62.0	5.8
ChAI	85.4	1.6	94.1	1.5	100.4	0.7	69.5	6.6	65.7	13.1	56.4	5.8
EA	91.9	3.1	105.6	2.6	111.2	1.6	48.4	2.2	46.6	12.2	38.0	6.4
UA	100.4	4.5	107.5	6.4	111.1	4.0	127.8	4.1	98.6	20.2	70.3	16.8
An	94.6	6.9	102.5	5.9	106.7	5.5	101.8	3.0	93.1	14.5	76.6	9.1

3.2. Method validation for five analytes in sanitary napkin and mouthwash samples

The specificity of the analytical method for sanitary napkins was assessed by comparing the analyses of matrix extracts with and without spiking the standard solution at the corresponding final concentration of the lowest concentration of the calibration curve. No interference was observed near the retention time of the five analytes and the IS in the unscented sanitary napkin extracts. The matrix effect was assessed at concentrations of 2, 6, and 15 ng/mL for atranol and 10, 30, and 75 ng/mL for the other analytes. The matrix effect values were calculated as $[\text{ME} (\%) = (\text{peak area of analyte spiked into matrix extract} / \text{peak area of analyte spiked into solvent}) \times 100]$ and ranged from 85.4 to 111.2 %, as shown in Table 2. The LOD and LOQ values calculated for the sanitary napkin matrix were 2.1–9.8 ng/g and 6.4–29.6 ng/g, respectively

(Table 3). The lowest concentration of each analyte in the calibration curve was determined based on the peak height required for unambiguous identification of the analyte on the UHPLC-MS/MS chromatogram.

Matrix-matched calibration curves were constructed using five different concentrations (10, 20, 50, 100, and 200 ng/g for atranol and 50, 100, 250, 500, and 1000 ng/g for the other four compounds), and excellent linearity was obtained ($r^2 > 0.9988$) (Table 3). Typical chromatograms of the matrix-matched calibration curve sample of sanitary napkins are shown in Fig. 3 (A). The accuracy and precision results are shown as the recovery (%) and relative standard deviation (RSD, %), which were obtained from analyzing three samples of different concentrations. The mean recoveries and RSDs were 86–108.4 % and 0.4–5.2 % for intra-day assays and 88.1–109.8 % and 2.1–11.5 % for inter-day assays, respectively (Table 5). These

Table 3. Linearity, limit of detection (LOD), and limit of quantification (LOQ) for the five allergenic compounds in a sanitary napkin

Chemicals	Range (ng) ^a	Regression equation				r ²	LOD (ng/g) ^b	LOQ (ng/g) ^b
		Slope		y-intercept				
		Mean	S.D.	Mean	S.D.			
Al	1-20	0.5403	0.0353	0.0081	0.0100	0.9997	2.1	6.4
ChAl	5-100	0.4524	0.0221	0.1183	0.0242	0.9988	3.7	11.3
EA	5-100	0.4666	0.0720	-0.1465	0.0268	0.9998	9.8	29.6
UA	5-100	0.3120	0.0674	-0.0453	0.0153	0.9988	9.5	28.8
An	5-100	0.1437	0.0270	-0.0850	0.2343	0.9990	5.8	17.5

a) ng in 0.1 g sanitary napkin. b) LOD and LOQ are expressed as the analyte concentration in the matrix (ng/g).

Table 4. Linearity, limit of detection (LOD), and limit of quantification (LOQ) for the five allergenic compounds in a mouthwash

Chemicals	Range (ng) ^a	Regression equation				r ²	LOD (ng/mL) ^b	LOQ (ng/mL) ^b
		Slope		y-intercept				
		Mean	S.D.	Mean	S.D.			
Al	1.5-100	0.0322	0.0041	0.0107	0.0120	0.9980	0.29	0.87
ChAl	1.5-100	0.0211	0.0025	-0.0026	0.0031	0.9992	0.40	1.21
EA	1.5-100	0.0181	0.0025	-0.0039	0.0023	0.9988	0.48	1.45
UA	1.5-100	0.0834	0.0136	0.0317	0.0129	0.9924	0.40	1.21
An	1.5-100	0.0301	0.0086	0.0095	0.0066	0.9958	0.43	1.31

a) ng in 0.5 mL mouthwash. b) LOD and LOQ are expressed as the analyte concentration in the matrix (ng/mL).

results are acceptable according to the criteria of accuracy and precision reported in the European SANTE/11813/2017 guidelines (70-120 % recovery and ≤ 20 % RSD).

Method validation for mouthwash analysis was performed using the as-prepared matrix, wherein four main ingredients of mouthwash samples (ethanol, glycerin, xylitol, and hydrogenated castor oil) were mixed. The validation procedure for this method was identical to that for sanitary napkins. No interference peaks were observed when the matrix extracts with and without the spiking of analytes were compared, indicating that this method guaranteed specificity for the determination of the five allergenic compounds in mouthwashes. The ME was assessed at concentrations of 5, 30, and 75 ng/mL for the five analytes. The ME (%) values were 38-127.8 % (Table 2), indicating that a matrix-matched calibration curve should be conducted for the accurate measurement of the five analytes in mouthwash. The LODs and LOQs for the mouthwash matrix were 0.29-0.48 and

0.87-1.45 ng/mL, respectively. Matrix-matched calibration curves were constructed using seven different concentrations (3, 4, 10, 20, 50, 100, and 200 ng/mL), and the determination coefficient (r^2) values exceeded 0.9924 (Table 4). Typical chromatograms of the matrix-matched calibration curve sample of the mouthwash are shown in Fig. 3(B). The mean recoveries and RSDs were 78.0-100.0 % and 0.6-7.3 % for intra-day assays and 87.8-107.2 % and 3.5-11.3 % for inter-day assays, respectively (Table 6). These results also fit the tolerance ranges of accuracy and precision (European SANTE/11813/2017 guidelines).

The validation results demonstrate that the two methods developed in this study are suitable for determining the five allergenic compounds commonly present in sanitary napkins and mouthwashes. The LODs and LOQs of our methods could not be directly compared with those of previously reported methods because, to the best of our knowledge, no studies have determined these compounds in sanitary napkins or mouthwashes. The LOQs of the LC-MS/

Table 5. Accuracy and precision for the five allergenic compounds in a sanitary napkin

Chemicals	Spiked (ng/g)	Intra-day (%)		Inter-day (%)	
		Recovery ^a	RSD	Recovery ^a	RSD
Al	20	101.5	2.6	103.1	4.3
	60	107.6	0.4	108.9	2.7
	150	102.3	2.7	106.7	7.6
ChAl	100	96.0	0.9	98.0	2.2
	300	102.1	2.0	102.7	2.1
	750	97.7	1.5	100.9	6.0
EA	100	95.8	4.1	96.7	3.0
	300	101.0	3.7	98.8	4.4
	750	104.0	1.0	102.2	3.3
UA	100	103.5	2.1	105.7	3.3
	300	108.4	2.1	109.8	3.0
	750	107.1	2.2	108.4	3.0
An	100	101.2	5.2	95.3	11.5
	300	99.2	4.1	95.7	7.3
	750	86.0	0.9	88.1	7.2

a) Mean of recovery from triplicate analysis.

Table 6. Accuracy and precision for the five allergenic compounds in a mouthwash

Chemicals	Spiked (ng/mL)	Intra-day (%)		Inter-day (%)	
		Recovery ^a	RSD	Recovery ^a	RSD
Al	10	78.0	2.8	90.4	10.5
	60	87.8	1.5	93.0	6.4
	150	81.1	1.2	91.5	10.7
ChAl	10	94.2	1.4	97.7	4.1
	60	89.6	0.6	94.3	6.1
	150	83.6	1.8	93.5	9.7
EA	10	91.0	1.4	92.6	7.8
	60	87.6	0.7	87.8	3.5
	150	85.3	4.8	90.9	7.5
UA	10	94.4	7.3	102.5	9.4
	60	100.0	4.6	107.2	7.0
	150	86.3	5.0	96.6	10.7
An	10	92.4	4.1	99.1	9.4
	60	93.6	4.9	100.7	8.6
	150	83.2	3.6	93.8	11.3

a) Mean of recovery from triplicate analysis.

MS method for the quantification of atranol and chloroatranol in perfume were 8.1 and 4.0 ng/mL,²¹ which were comparable to those of our methods (Table 3, 4). The LOQs of the LC-MS/MS method for atranol and chloroatranol in moss extracts²⁰ and

atranorin and usnic acid in lichen extracts¹⁹ were 1.1-1.5 µg/mL and 41.0-212.9 ng/mL, respectively. Compared with other methods that determine allergenic compounds in perfume and lichen (moss) extracts, the two methods established in this study exhibited

sufficient sensitivity (ppb level) to quantify allergenic compounds present in personal hygiene products.

3.3. Monitoring allergenic compounds in sanitary napkin and mouthwash samples

The established and validated methods were employed to analyze the presence of the five allergenic compounds in commercially available sanitary napkins and mouthwashes. A total of twelve sanitary napkin brands and fifteen mouthwash types were purchased from retail stores and processed in accordance with the sample pretreatment protocol described earlier. Despite the methods being capable of detecting compounds at the parts-per-billion (ppb) level, none of the five allergenic compounds were detected in the analyzed commercial products. However, due to the potential of personal hygiene products to cause allergic contact dermatitis,⁵⁻¹⁰ long-term monitoring is essential. It is considered that the proposed method, which enables simultaneous and sensitive detection of the five allergenic compounds in commercially available products, can facilitate such monitoring efforts.

4. Conclusions

The aim of this study was to develop highly sensitive analytical methods that can simultaneously detect the five allergenic compounds in sanitary napkins and mouthwashes. Optimization of the mobile phase composition of UHPLC-MS/MS allows highly sensitive detection of analytes. The methods described in this study were validated in compliance with the ICH Q2(R1) and European SANTE/11813/2017 guidelines and were satisfactory in terms of specificity, LOD, LOQ, linearity, accuracy, and precision. To the best of our knowledge, no studies have simultaneously determined five allergenic compounds in sanitary napkins or mouthwashes. The proposed method can be applied for the determination of allergenic compounds in other personal hygiene products.

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