Determination of cyclic volatile methylsiloxanes in personal care products by gas chromatography

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Abstract

OBJECTIVE: Organosiloxanes are prevalent in personal care products (PCPs) due to the desired properties they impart in the usage and application of such products. However, the European Chemical Agency (ECHA) has recently published restriction proposals on the amount of two cyclic siloxanes, octamethylcyclotetrasiloxane (D4) and decamethylcyclotetrasiloxane (D5), allowed in wash off products such as shampoos and conditioners which are discharged down the drain during consumer use. This legislation will require that reliable analytical methods are available for manufacturers and government agencies to use in documenting compliance with the restrictions. This article proposes a simple analytical method to enable accurate measurement of these compounds down to the circa 0.1 weight per cent level in PCPs.

METHODS: Although gas chromatography methods are reported in the literature for quantitation of D4 and D5 in several matrices including PCPs, the potential for generation of false positives due to contamination, co-elution and in situ generation of cyclic volatile methylsiloxanes (cVMS) is always present and needs to be controlled. This report demonstrates the applicability of using a combination of emulsion break, liquid-liquid extraction and silylation sample preparation followed by GC-FID analysis as a suitable means of analysing PCPs for specific cVMS.

RESULTS: The reliability and limitations of such methodology were demonstrated through several round-robin studies conducted in the laboratories of a consortium of silicone manufacturers. In addition, this report presents examples of false positives encountered during development of the method and presents a comparative analysis between this method and a published QuEChERS sample preparation procedure to illustrate the potential for generation of false positives when an inappropriate approach is applied to determination of cVMS in personal care products.

CONCLUSION: This report demonstrates that an approach to determine cVMS levels in personal care products is to perform an emulsion break on the sample, isolate the non-polar phase from the emulsion break and treat with a silylation reagent to abate potential in situ formation of cycles during the course of GC-FID analysis. Round-robin studies conducted in laboratories representing multiple siloxane manufacturers demonstrated the reliability of the GC-FID method when measuring cVMS in PCPs down to circa 0.1%.

Résumé

OBJECTIF: Les organosiloxanes sont des composés utilisés essentiellement dans les produits de soins corporels (PSC) en raison des propriétés recherchées qu'ils confèrent quant à l'utilisation et application de tels produits. Cependant, l'Agence européenne des produits chimiques (AEPC) a récemment émis des propositions de restriction en quantité pour deux des siloxanes cycliques – l'octaméthylcyclotétrasiloxane (D4) et le décaméthylcyclotétrasiloxane (D5) – qui sont autorisés dans les produits cosmétiques de type « wash off » tels que les shampoings et préparateurs capillaires et qui sont habituellement évacués de la surface du corps par rinçage à l'eau (sous la douche) durant leurs utilisations. Cette législation impliquera que des méthodes d’analyses fiables soient disponibles chez les producteurs et les agences gouvernementales afin de documenter le respect de ces restrictions. Cette publication propose une méthode analytique simple pour permettre une mesure précise de ces composés pour des teneurs aussi faibles que 0.1% en poids dans les produits cosmétiques.

MÉTHODES: bien que la chromatographie en phase gazeuse soit une méthode décrite dans la littérature scientifique pour la quantification de D4 et D5 dans de nombreuses matrices en ce inclus les PSC, la probabilité d’obtenir de faux positifs en raison de la contamination, la co-elution et la création in situ de méthylsiloxanes volatiles cycliques (MVC) est toujours présente et doit être contrôlée. Ce rapport démontre l’adéquation de la combinaison d’anti-émulsions, d’extraction liquide-liquide et de la préparation de l’échantillon via silylation, suivi de GC-FID comme technique convenable pour analyser les PSC par rapport aux MVC.

RÉSULTATS: la fiabilité et les limitations de cette méthodologie ont été démontrées par une étude expérimentale comparative dans les laboratoires d’un consortium de producteurs des silicones. De plus ce rapport présente des exemples de faux positifs rencontrés durant le développement de la méthode et met en lumière, via une étude...

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comparative par rapport à la méthode publiée QuEChERS de préparation d’échantillons, le potentiel pour obtenir de faux positifs lorsqu’une approche inappropriée est utilisée durant la mesure des MVC présents dans les PSC.

**CONCLUSION:** Ce rapport démontre qu’une approche pour déterminer la teneur en MVC dans les produits cosmétiques consiste à appliquer des méthodes anti-emulsion sur l’échantillon, d’isoler la phase apolaire de la partie non-emulsifiée et de la traiter avec un agent de silylation afin d’éviter la potentielle production de MVC in situ durant l’analyse CG-FID. Une étude expérimentale comparative en laboratoire menée par plusieurs fabricants de composés siloxanes a démontré la fiabilité de la méthode CG-FID pour mesurer les teneurs en MVC dans les PSC aussi faibles que 0.1% en poids.

**Introduction**

Silicones have wide application and utility in a variety of materials and products due to their unique properties. One particular area where they are widely used as additives are in many different types of personal and beauty care products including shampoos, conditioners, hair care products, body lotions and sun protection formulations. Addition of silicones to these types of product formulations imparts specific attributes such as ease of application, desirable sensory properties and wash off resistance [1]. Personal care products with general silicone ingredient names such as dimethicone or cyclomethicone, or more specific names such listed such as cyclopane-tasiloxane, indicate that these materials have been intentionally added as a component to the formulation. Although the silicones used may contain specific functional groups, or may be a copolymer (e.g. silicone polyethers) to bring specific properties or attributes to a formulation, the most prominent type of functional group is the SiO2 repeat unit. A product or formulation that contains polydimethylsiloxane (PDMS) may have residual cyclic dimethyl siloxane oligomers present as well. Accurate quantitation of cyclic siloxanes in personal care products is important to support studies and assessments that require this information.

In silicone chemistry nomenclature, hexamethyldisiloxane (CAS 541-05-9), octamethyldisiloxane (CAS 556-67-2), decamethyldisiloxane (CAS 541-02-6) and dodecamethyldisiloxane (CAS 540-97-6) are often referred to as D3, D4, D5 and D6, respectively, where D is the Me2SiO2/2 repeat unit and the numeral following designates the number of repeat units. Such compounds are also commonly categorized as cyclic volatile methyl siloxanes (cVMS). The chemical structures of D4, D5 and D6 are shown in Fig. 1.

Personal care products are often complex formulations that contain many components that can complicate accurate analysis of cyclic siloxane content. If levels of the analytes are very low, considerations and concerns regarding contamination of samples from analysts, instruments and equipment must be taken into account [2, 3]. In addition, if the sample contains siloxane polymer as an ingredient, the possibility of generation of additional cVMS compounds from degradation of siloxane polymer during the course of analysis requires consideration. Potential causes of inaccuracy have been described in the analysis of silicone emulsions [4] but have not been sufficiently addressed in the analysis of personal care products.

There are not many examples in the literature regarding analysis of cVMS siloxanes in personal care products. A few studies have attempted to assess the quantity of cVMS compounds in personal care products to use in dermal exposure and wash off calculations [5–9]. However, analytical results and observations reported in these studies seem to indicate that, in some cases, quantitative results reported may include contributions from artefacts and may not reflect the actual level of cVMS compounds in the samples analysed [5, 6]. The purpose of this article was to propose a simple GC-FID method to enable accurate measurement of cVMS compounds down to the circa 0.1 weight per cent level in personal care products that contain silicones as additives. In addition, the potential for pitfalls and generation of artefacts in the absence of suitable methodology is discussed.

**Materials and methods**

**GC method**

Multiple laboratories participated in the development and assessment of the described GC method through round-robin studies. Materials and chemicals for the GC method were sourced locally by each of the laboratories participating in the study. Sources used by one of the laboratories are shown for example, but differed for other members participating in the study.

**Chemicals**

Octamethylocyclotetrasiloxane (D4), decamethylocyclopentasiloxane (D5) and dodecamethylocyclohexasiloxane (D6) were obtained from Wacker, Aldrich and ABCR, respectively. Normal dodecane was used as an internal standard and was obtained from Fluka. Hexane, acetonitrile and N,N-dimethylacetamide were obtained from Merck, Aldrich and Scharlau, respectively. N-methyl, N-trimethylsilyl trifluoroacetamide (MSTFA) was obtained from Machery-Nagel.

**Materials**

A Satorius Analytical balance (0.1 mg increments) was used for measurement of sample weights. Screw-cap glass vials (15 mL) and headspace vials with septum crimp caps (20 mL) were obtained from Supelco and PerkinElmer. A Heidolph vortex shaker was used for mixing samples. Standards were prepared using a combination of Brand volumetric flasks, Eppendorf adjustable pipettes and Hamilton gas tight microfille syringes.

**Gas chromatograph**

An Agilent 6890 GC equipped with a split/splitless injector was used and was equipped with an injection splitter for mounting of two columns. An HP-1 and HP-5 30 m × 0.32 mm × 0.25 μm column from Agilent was used. All participants in the study
typically used HP-1 or HP-5 type stationary phases of generally similar dimensions. A temperature program of 60–150°C @ 8°C min⁻¹ was used for separation followed by a thermal ramp at 25°C min⁻¹ to 300°C for 5 min. A flame ionization detector at 300°C was commonly used. Helium was used as the carrier gas at 2 mL min⁻¹ constant flow. Other testing sites used similar conditions, but actual parameters such as column types and temperature programs did vary due to differences among instrument configurations and established workflows at each of the sites participating in the round-robin study.

GC method and sample preparation

An internal standard solvent solution was prepared by placing 400 mg of n-dodecane into a 200-mL volumetric flask. Subsequently, 50 mL of dimethylacetamide was added and the solution was diluted to the mark with acetonitrile. A 400-mg sample weighed to the nearest 0.1 mg was placed into a 15-mL screw-cap glass and treated with 2 mL of the of 75/25 acetone/dimethylacetamide internal standard solution and dispersed by gentle shaking. Next, 8 mL of hexane was added, and the closed vial was vigorously shaken on a vortex mixer for one minute. After phase separation, 1 mL of the upper hexane phase was transferred into a GC autosampler vial and treated with 100 μL of MSTFA. The mixture was then incubated for 30 min at 80°C. A one microlitre aliquot was used for GC analysis. This sample preparation followed by GC-FID analysis on a non-polar stationary phase wall coated open tube capillary column was the main method used in the inter-company round-robin studies for the analysis of personal care product formulation and commercial samples.

Two blank runs were performed for each set of samples and were prepared in the same way without any sample present. The blank samples are used to verify day-to-day applicability of the GC system for the analyses being conducted.

Calibration

Calibration solutions were prepared by weighing 100 mg of each cVMS (D4, D5, D6) into a 10-mL volumetric flask and diluting to the mark with hexane to form stock solution A. Exactly 1 mL of this stock solution was diluted with hexane in another 10 mL volumetric flask to form stock solution B. An internal standard calibration solution was prepared by weighing 80 mg of n-dodecane into a 100-mL volumetric flask and diluting to the mark with hexane. Ten calibration solutions were then prepared in separate 10 mL volumetric flasks. Exactly 5 mL of the internal standard solution was added to each flask. Subsequently, 25, 50, 100, 150 and 200 μL of stock solution B and 50, 100, 200, 500 and 1000 μL of stock solution A are added to one of the volumetric flasks using different sizes of microlitre syringes and diluted to the mark with hexane. The calibration solutions are stable over a period of at least 2 weeks when stored in a refrigerator. Least squares simple linear regression was used to determine the experimental response factor relative to the internal standard for each of the cVMS analytes.

Inter-laboratory round-robin studies

Five different laboratories (A, B, C, D, E) performed the sample preparation and GC-FID method as described above in round-robin studies. Initial round-robin studies were performed with laboratory formulations of shampoo and conditioners. An additional round-robin study was performed with several types of commercially available personal care products.

Preparation of shampoo and conditioner lab formulation samples

Three shampoo and three hair conditioner formulations (Table SI, supporting information) were prepared in the laboratory for evaluation of the GC-FID method used in inter-company round-robin studies. The formulations were designed to reflect the types of ingredients that may be found in personal care products. For both the shampoo and conditioner, one formulation was prepared with no added silicone, another with an added silicone ingredient, and another spiked with known levels of cyclic siloxanes. An additional shampoo formulation that contained dimethiconol as an additive and a conditioner formulation that contained amodimethicone and dimethiconol (Table S2, supporting information) were also prepared in the laboratory for evaluation of the GC method used in the inter-company round-robin studies.

Commercially available personal care product samples

Commercially available conditioners, shampoos and body lotions were sourced in Europe. The list of samples analysed in the round-robin studies are shown in Table I.

Additional sample preparations and analysis by GC-MS

Auxiliary experiments involved the use of an additional sample purification step using solid phase extraction. In these experiments, 1 mL of the hexane extract obtained from the main sample preparation procedure was purified by micro-column chromatography by placing the aliquot on a stationary phase bed that contained 200 mg of sodium sulphate and 800 mg of Florisil. The extract was eluted with 5 mL of 99/1 hexane/methyl tertiary butyl ether by directly pouring over the SPE cartridge. The first 2 mL was sent to waste, and the fraction from 2 to 4 mL elution volume was used for analysis. These samples were analysed by single ion monitoring GC-MS using ¹³D₄ as the internal standard.

Results and discussion

Gas chromatography is a useful technique for the determination of cyclic siloxanes in materials, including personal care products. The

Table I Commercially available personal care products purchased off of the shelf. Body lotion, conditioner and shampoo samples are designated as BL, CO, SH, respectively.

| Sample | Silicone component |
|--------|--------------------|
| BL1    | None added         |
| BL2    | PDMS methyl end-capped |
| CO1    | None added         |
| CO2    | PDMS (OH end-capped) |
| CO3    | Silicone quat, PDMS (methyl end-capped) |
| CO4    | D5                 |
| SH1    | None added         |
| SH2    | PDMS (methyl end-capped) |
The purpose of this publication is to recommend a GC-FID method to simply and reliably determine cVMS in personal care products down to the circa 0.1% level, and to also demonstrate what precautions and considerations need to be taken to prevent the generation of artefacts which can result in false positives or artificially high results when performing analyses of this type, especially when attempting analyses at lower concentration levels.

Round-robin analysis of laboratory formulations and commercially available personal care products

Several silicone producers participated in inter-company round-robin studies to assess the applicability and ruggedness of a GC-FID method for determination of cVMS in several types of personal care products. Analyses were performed on both laboratory formulations (supporting information, Tables S1 and SII) and off the shelf commercially available products (Table I). Analysis of laboratory formulations allowed complete knowledge of the sample matrix thereby removing uncertainty associated with sample composition. In summary, sample preparation involved treatment with a combination of a polar solvent mixture (acetonitrile/dimethylacetamide) and immiscible non-polar solvent (hexane) to break the product emulsion. The cVMS compounds preferentially partition into the non-polar phase and are separated from polar components in the sample. An aliquot of the non-polar hexane phase is then treated with MSTFA as a silylation reagent and then analysed by GC-FID.

Results for D4, D5 and D6 levels from the round-robin study of two different sets of laboratory formulations are shown in Tables II and III, and results from analysis of several commercially available personal care products are shown in Table IV. Overall, the inter-laboratory results were in generally good agreement, especially at the higher concentration levels. Averages and standard deviations were calculated for a sample if more than one laboratory detected a result above background. Results obtained from the analysis of both the shampoo and conditioner formulations spiked at the 500 ppm (0.05%) level exhibited the highest reproducibility among the samples analysed in Table II. The %RSD values were 16% or less. Generally, the limit of detection for these analyses conducted with GC-FID instrumentation was on the order of 10–50 ppm and varied somewhat across the laboratories due to instrumentation configuration and condition differences that would be expected in an inter-laboratory study. An apparently artificially high result for D4 was detected by laboratory B when the shampoo 2 sample in Table II was analysed on one type of column (HP-1), but was not detected when analysed on a column with different selectivity (HP-5). Another artificially high result was that for D6 in the analysis of conditioner 2 in Table II by laboratory C. The detection of false positives in some of the results points out the need for caution when analysing for low level cyclic siloxanes in materials. Instrument components can be a source of contamination, and cross sample contamination from analysis of other siloxane samples using the same equipment may also be a source of interfering background signals. When performing analyses using non-selective detectors such as an FID, a false positive can also result from co-elution of the cyclic siloxane with another compound in the sample. It is highly recommended that confirmatory analyses be performed to obtain accurate results, especially if a complex matrix and resultant complex chromatograms clearly indicate interference issues, and that method blanks be included to account for system contributions that may generate false positives.

| Table II | Inter-company round-robin results (ppm) using GC-FID for determination of cVMS analytes in laboratory formulations of shampoos and conditioners (described in Table S1) with and without added silicone. Values shown in bold and underlined are artificially high results. Different laboratories are designated as A, B, C, D and E. ND indicates no detection of signal. A value reported as less than a quantity indicates detection of a signal below the limit of quantitation. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Shampoo 1 (no silicone) | Shampoo 2 (silicone added) | Shampoo 3 (silicone plus cVMS spike) |
| Lab | D4 | D5 | D6 | D4 | D5 | D6 | D4 | D5 | D6 |
| A | ND | ND | <20 | ND | 10 | <50 | 480 | 500 | 510 |
| B (HP5)* | <10 | <10 | <10 | <10 | 10 | 20 | 540 | 490 | 450 |
| B (HP1)* | 20 | 20 | 10 | 100 | 30 | 10 | 600 | 490 | 440 |
| C | <10 | <10 | <10 | <10 | 14 | 21 | 439 | 471 | 453 |
| D | <10 | 25 | <10 | 10 | 38 | 17 | 454 | 436 | 422 |
| E | 7 | 12 | 2 | 12 | 30 | 17 | 525 | 565 | 520 |
| Average | 14 | 19 | 7 | 42 | 26 | 16 | 506 | 509 | 471 |
| st. dev. | 9.2 | 6.6 | 3.8 | 60 | 35 | 35 | 7.4 |
| % RSD | 68 | 35 | 471 |

| | Conditioner 1 (no silicone) | Conditioner 2 (silicone added) | Conditioner 3 (silicone plus cVMS spike) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Lab | D4 | D5 | D6 | D4 | D5 | D6 | D4 | D5 | D6 |
| A | ND | ND | <20 | 30 | 10 | 10 | 480 | 480 | 470 |
| B (HP5)* | <10 | <10 | <10 | 20 | 10 | 20 | 500 | 470 | 460 |
| B (HP1)* | 10 | 20 | 10 | 30 | 30 | 10 | 520 | 460 | 460 |
| C | <10 | <10 | <10 | 37 | 15 | 233 | 471 | 458 | 462 |
| D | <10 | 17 | <10 | 17 | 33 | 16 | 455 | 504 | 451 |
| E | 8 | 16 | ND | 33 | 27 | 20 | 513 | 528 | 495 |
| Average | 9 | 18 | 1.4 | 28 | 21 | 52 | 490 | 483 | 501 |
| st. dev. | 1.4 | 2.1 | 7.7 | 10 | 89 | 25 | 27 | 80 |
| % RSD | 16 | 12 | 52 |

*Analysis on different stationary phases.

Results from analysis of laboratory formulations that contain dimethiconol (PDMS with silanol termini) and amodimethicone (PDMS with amino functionality) are shown in Table III. Overall, good agreement was obtained in the results obtained from the analyses conducted in the different laboratories. A 5% RSD was observed for D4 when present at ~0.02% and a 5–10% RSD was observed for D5 and D6 when present at ~0.005%. Laboratory E also measured the D4 content in both formulations in Table III by silicon-29 NMR. The results were in very good agreement with the GC-FID results. The shampoo was found to contain 194 ppm D4 by NMR and 200 ppm D4 by GC-FID analysis. The conditioner was found to contain 27 ppm D4 by NMR and ~50 ppm D4 using split injection GC-FID and 30 ppm D4 using cool on column injection GC-FID. Laboratory E analysed 10 replicates of both formulations and determined an intra-laboratory short-term repeatability less than 15%RSD for all analytes.

The GC-FID method was also applied to analysis of off the shelf personal care products (Table I). Most of the personal care products analysed indicated very low (near the detection limit) or non-
detected cVMS content (Table IV). The BL1 sample had no indication on its ingredients label that silicone was present in the formulation. The BL2 sample ingredient list indicated the presence of methyl end-capped PDMS (dimethicone) in the formulation. However, D4, D5 and D6 were generally not detected in the BL2 sample above the estimated limit of detection (ca. 10 ppm) for analyses conducted using the GC-FID method.

The CO1 sample had no indication on its ingredients label that silicone was present in the formulation and all laboratories indicated levels near or below the general detection limit except the D4 value from laboratories C and D which appear to be false positives. The CO2 sample label indicated the presence of OH-ended silicones (dimethiconol) in the formulation, and all of the laboratories identified the presence of D4 at nearly consistent levels. The CO3 sample label indicated the presence of silicone quaternary material (silicone quaternium-22) and trimethyl-ended PDMS (dimethicone). Only very low levels of cVMS were found in this sample, generally near or below the detection limit for all of the analytes. The CO4 label indicated the presence of D5 (cyclopentasiloxane) in the formulation. All laboratories found D5 in CO4 at nearly consistent levels (>0.5%, 8% RSD), with lower levels of D4 and D6 present. The SH1 shampoo sample does not contain any silicones according to ingredient label information on the product, and the majority of analyses conducted in round-robin testing did not detect the presence of cVMS. The SH2 sample did contain trimethyl-ended PDMS (dimethicone) according to the product label. The majority of analyses conducted indicated cVMS content in the sample near or below the general detection limit.

Considerations regarding analysis of cVMS in personal care products

Personal care products are usually complex multi-component formulations that may contain water, surfactants, perfumes, stabilizers, preservatives, antioxidants, salts and other types of molecules and polymers including silicones. Personal care products may contain different types of silicones, including cyclic, linear and organofunctional PDMS with the choice dictated by the desired benefit or attribute. Cyclic silicones have been used as volatile carriers for antiperspirants, amino functional polymers have been used in rinse-off conditioners, and silicone emulsions have been used in two-in-one conditioning shampoo [1]. Silicones in personal care products may range from low molecular weight molecules to high molecular weight molecules containing different types of silicones, including cyclic, linear and organofunctional PDMS with the choice dictated by the desired benefit or attribute.

Cyclic siloxanes analysis personal care products

Table III Inter-company round-robin results (ppm) using GC-FID for determination of cVMS analytes in laboratory formulations (described in Table SII) of shampoos and conditioners that contain dimethiconol and amodimethicone. Different laboratories are designated with A, B, C, D and E.

| Lab | D4 | D5 | D6 | D4 | D5 | D6 |
|-----|----|----|----|----|----|----|
| A   | 166| 57 | 52 | 30 | 17 | 57 |
| B   | 170| 58 | 44 | 33 | 17 | 59 |
| C   | 179| 73 | 39 | 45 | 29 | 54 |
| D   | 173| 57 | 44 | 30 | 14 | 51 |
| E   | 200|70 | -60| -60| -60| -60 |
| E*  | 205|69 | 42 | 30 | 19 | 59 |

Table IV Inter-company round-robin results of cVMS levels (in ppm) in commercially available body lotion, conditioner and shampoo samples determined by emulsion break liquid-liquid extraction followed by GC-FID analysis. Different laboratories are designated with A, B, C, D and E. ND indicates no detection of signal. A value reported as less than a quantity indicates detection of a signal below the limit of quantitation.

| Lab       | D4 | D5 | D6 | D4 | D5 | D6 |
|-----------|----|----|----|----|----|----|
| BL1 (no silicone) | ND | ND | ND | 163| 43 | 22 |
| BL2 (silicone ingredient) | 18| 9 | 10 | 176| 80 | 17 |
| C         | 145|35 | ND | 190|59 | 22 |
| D         | 75 | <10<10<10| 181|87 | 23 |
| E         | ND | ND | ND | 250|ND | ND |
| Average   | 79 | 22 | 192|67 | 21 |
| st. dev.  | 64 | 18 | 34 | 20 | 2.7 |
| % RSD     | 80| 82 | 18 | 30| 13 |

| Lab       | D4 | D5 | D6 | D4 | D5 | D6 |
|-----------|----|----|----|----|----|----|
| CO1 (no silicone) | 14| 10| 10 | 34| 5150| 16 |
| CO2 (silicone ingredient) | 16|10 |10 | 36|5225 | 69 |
| C         | 26 | 22 | 10 | 40|6266 | 18 |
| D         | <10<10<10<10| 20|5230 | 19 |
| Average   | 19 | 16 | 10 | 33|5474 | 31 |
| st. dev.  | 6.4|8.5| 0.0| 8.7|462 | 26 |
| % RSD     | 34| 53 | 0.0| 27|8.4 | 84 |

| Lab       | D4 | D5 | D6 | D4 | D5 | D6 |
|-----------|----|----|----|----|----|----|
| SH1 (no silicone) | ND| ND| ND | <10|10 | 10 |
| SH2 (silicone ingredient) | ND| ND| ND | 12|<10 | 13 |
| C         | ND | ND | ND | 16|25 | 14 |
| D         | <10<10<10<10| <10|<10 | 18 |
| E         | ND | ND | ND | ND|ND | ND |
| Average   | 14 | 18 | 14 |
| st. dev.  | 2.8| 11 | 3.3 |
| % RSD     | 20| 61 | 24 |

*Analysis by GC-FID with cool on column injection instead of split injection.
molecular weight, high viscosity polymers. They also may be present in formulations as resin networks or dispersions of cross-linked elastomers. In addition, the end groups on the siloxane polymer chains may differ in functionality, where dimethicone may be, for example, a trimethylsilyl end-capped polymer, whereas dimethiconol has silanol end groups.

The contents of the matrix must be considered to determine a suitable approach for analysis. The absence of such considerations may result in use of an analytical approach that could yield inaccurate results. There are only a few reports in the literature describing methods of analysis for determination of cVMS compounds in personal care products [5–9]. In most cases, the analytical methodology reported is not the focus of the study. Although efforts are made in these published studies to take precautions to prevent siloxane contamination from analysts and the instrumentation (e.g., introduction of siloxanes from inlet and sample vial septa) as well as attempts to validate the methods used, there appears to be an absence of consideration for the composition and chemistry associated with the sample matrices under study.

Potential for thermal degradation in GC

Thermal degradation studies of siloxanes have shown that the polymers can degrade through a back biting mechanism where the silanol end group of the polymer chain can attack a silicon atom several repeat units back on the chain with formation of a cyclic siloxane oligomer and a shorter polymer chain as products [10]. A schematic representation of this type of degradation is shown in Fig. 2. Furthermore, degradation of polysiloxanes can be catalysed by the presence of acids or bases in the system. Injection of samples containing OH-terminal linear siloxanes (commonly listed as dimethiconol on personal care product ingredient lists) into the hot inlet of a gas chromatograph has the potential to form cyclic siloxanes. This is a known phenomenon [4, 11]. An approach to reduce this potential is to treat the sample preparation with a silylation reagent (such as MSTFA). The addition of the MSTFA can serve several purposes. It may modify reactive silanol groups and abate back biting of the siloxane, it may silylate other reactive species in formulations and reduce their potential to behave as catalysts in siloxane degradation, and it may derivatize co-extracted substances which might interfere with the chromatographic analysis.

Cool on column injection provides a possible option for avoiding issues associated with thermal degradation during the GC injection and vaporization process, and also potentially allows for lower limits of detection. Disadvantages for this type of injection, however, are increased column contamination compared to split injections, and cool on column injection hardware is not commonplace. For this reason, the authors decided to use a more common split injection GC-FID method in the round-robin studies so that the method could potentially be applied to a variety of laboratories for those interested in attempting similar analyses for determination of cVMS in personal care products.

Potential for interferences

Overall, generally consistent results were obtained from the different companies participating in the round-robin study when materials actually contained cVMS components at levels well above detection limits. In many instances where false positives were obtained in this study, reanalysis on a stationary phase with even slightly different selectivity clearly indicated misidentification in the original analysis.

Figure 2 Schematic of siloxane backbiting mechanism. The schematic shows formation of D₄ cyclic siloxane but other molecular weight species (e.g. D₃, D₅, D₆...) can also be formed from backbiting of the siloxane onto itself.

False positives were more frequent, and wider variation was observed when results were at concentration levels in the vicinity of the general detection limit (10–50 ppm) of the GC-FID method when applied in the different laboratories of the round-robin study. Such observations reiterate the need to confirm positive results by use of a confirmatory column, referee technique or analysis with a more specific detector (e.g. mass spectrometry). However, it is important to note that use of a mass spectrometer as a detector does not remove the potential for false positives due to siloxane contamination from system components or in situ generation of cVMS during the course of analyses from linear siloxane polymers that also may be present in the samples.

Impact of sample preparation on chromatography

The emulsion break, liquid–liquid extraction sample preparation procedure used for the round-robin experiments extracts polar constituents into the polar solvent and non-polar constituents (including the cVMS of interest) into the non-polar solvent. Only the non-polar phase is analysed by GC, so components removed from the sample by the polar solvent phase are not injected into the chromatograph. Removal of polar components from the sample simplifies the chromatography and reduces the chance for interference from co-eluting components and removes components that could potentially catalyse degradation of siloxane in the hot inlet of the GC. Ancillary experiments were also performed where solid phase extraction was added as an additional sample clean up step. An aliquot of the non-polar phase from the emulsion break was passed through a short Florisil column. The cVMS can be eluted from the column while other sample matrix components are retained. This results in a cleaner sample for analysis. The effectiveness of sample cleanup is qualitatively illustrated in Fig. 3 which compares GC-FID chromatograms from analysis of a shampoo sample when extracted with only a polar solvent (acetone, Fig. 3A) versus extraction with a combination of immiscible polar and non-polar solvents (Fig. 3B). The chromatogram from the latter preparation can be further simplified by addition of solid phase extraction to the sample preparation procedure (Fig. 3C and D).

Impact of sample preparation on false positives

The general importance of the sample preparation for the accurate analysis of cVMS in personal care products was demonstrated above. However, while sufficient chromatographic resolution and
verification of positive findings are essential prerequisites for assessing the reliability of the results, especially at low concentrations, sometimes even the chemistry itself in the sample preparation procedures must be critically examined.

Recently, published studies [5, 6] reported the use of a QuEChERS approach (QuEChERS refers to the “Quick, Easy, Cheap, Effective, Rugged and Safe” sample preparation procedure commonly used in pesticide analysis applications. See, for example, Ref. 12) in sample preparation for determination of cVMS analytes in a variety of personal care products. The results found in that study were surprising as D3 was detected at the same magnitude and frequency as D5, and unlike D5, D3 is usually not applied (and to the authors’ knowledge, never applied) as an ingredient in cosmetic formulations. The QuEChERS approach (developed for analysis of pesticide residues in food and agricultural products) uses high levels of magnesium sulphate and sodium acetate as well as a primary secondary amine (PSA) adsorbent and a C18 adsorbent in order to separate polar and slightly polar substances from non-polar analytes. The presence of D3 in most PCPs analysed was a strong indication that utilization of these materials in the sample preparation for cVMS analysis may influence the thermal degradation of silicone polymers in the samples to form additional cVMS during the course of analysis.

In a comparative laboratory study, the published procedure [6] was compared to a procedure based on the emulsion break, solvent extraction method used for preparation of samples in the round-robin studies. The comparison required lower detection limits, thus the emulsion break, solvent extraction method utilized GC-MS SIM detection and the application of a more rigorous sample preparation to reduce interferences. Extract cleanup by solid phase extraction step was performed by passing an aliquot of the hexane extract through a Florisil column prior to analysis as described in the materials and methods section. Quantification was performed by single ion monitoring GC-MS using $^{13}$D4 as an internal standard.

Both methods were applied to the body lotion (BL2) containing methyl end-capped PDMS and shampoo (SH1) which contained no silicone polymer. The results are summarized in Table V.

While all cVMS analytes D3, D4, D5 and D6 were detected in sample BL2 when analysed using the QuEChERS procedure, none of these cVMS analytes were detected with application of the emulsion break/extraction/SPE procedure. However, there were no detectable cVMS observed by either method for the analysis of Table V

**Levels of cVMS (in ppm) determined in commercially available body lotion and shampoo using published QuEChERS method versus preparation using an emulsion break, solvent extraction followed by SIM GC-MS analysis.**

| Product (type of silicone ingredient) | Method                          | D3  | D4  | D5  | D6  |
|--------------------------------------|---------------------------------|-----|-----|-----|-----|
| BL2 (dimethicone)                    | QuEChERS                        | 5   | 7   | 5   | 2   |
| SH1 (no silicone)                    | Emulsion break/extraction/SPE   | <0.2| <0.2| <0.2| <0.2|
| SH1 (no silicone)                    | QuEChERS                        | <0.2| <0.2| <0.2| <0.2|
| SH1 (no silicone)                    | Emulsion break/extraction/SPE   | <0.2| <0.2| <0.2| <0.2|

'<0.2' not detected up to the method detection limit MDL of the procedure.
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sample SH1 which did not contain silicone polymer as a listed ingredient. Both analyses were validated by recovery experiments. Good recoveries in the emulsion break/extraction/SPE method indicated that the lower cVMS results when using the emulsion break method were not due to unaccountable loss of analytes during sample preparation.

The results of this comparative laboratory experiment demonstrate that sample preparations like QuEChERS, even if developed especially for the analysis of non-polar analytes, may not be a suitable sample preparation technique for determination of cVMS in personal care products. A potential rationale for this observation is that the high concentration of salts and ionic adsorbents used in the QuEChERS sample preparation in combination with surfactants and emulsifiers common in personal care product formulations may aid in transfer of salts and other substances into the hexane extract, which then influence the thermal degradation of silicone polymers during the GC analysis.

Conclusion

Overall, GC is a suitable technique for determination of cVMS in a variety of matrices and has been well documented in the literature. Recent studies have been published regarding the measurement of cVMS in personal care products, but results have indicated possible concerns regarding the accuracy of such measurements due to issues such as formation of cVMS during the course of analysis and contamination of instrumentation with non-volatile components which can exacerbate the problem of inaccurate quantitation or the generation of false positives. Appropriate sample preparation procedures as well as analysis of appropriate controls or method blanks are required for accurate quantitation of cVMS in personal care products. This report demonstrates that one approach to determine cVMS levels in personal care products is to perform an emulsion break on the sample, isolate the non-polar phase from the emulsion break and treat with a silylation reagent to abate potential formation of cyclics during the course of analysis (specifically, in the hot inlet of the GC). Round-robin studies were conducted in laboratories representing multiple silicone manufacturers to demonstrate the reliability of such an approach to analysis when measuring cVMS in personal care products down to the circa 0.1% concentration level. The emulsion break procedure followed by GC-FID analysis is a recommended approach to analysis given its relative simplicity and broad applicability to a variety of laboratory settings. However, the round-robin studies conducted among different laboratories also demonstrated that false positives can occur due to system contamination or co-elution with other components extracted from the sample, which tend to become more problematic and pronounced at lower concentration levels. Laboratories may experience contamination issues with introduction of non-volatile siloxanes to the inlet of the GC through analysis of multiple samples in a campaign. Such non-volatile siloxane residues have the potential to generate cyclic siloxanes during an injection process, especially if a species that catalyses degradation of siloxanes into cyclic siloxanes is injected. Method blanks and confirmatory analyses are highly recommended, especially in those situations that indicate complicated chromatography. In some instances, more selective detectors, such as a mass spectrometer, or additional sample preparation procedures (e.g. solid phase extraction) may be required. Furthermore, care and consideration must be given to sample preparation procedures that can potentially exacerbate or catalyse decomposition of siloxanes in samples to form cVMS compounds not originally present in the sample. As an example, the use of QuEChERS, which is mainly designed for the analysis of pesticide residue in foods, is not recommended in the preparation of personal care product samples for cVMS analysis.

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References

1. Andriot, M., DeGroot, J.V. Jr, Meeks, R. et al. Silicones in industrial applications. In: Inorganic Polymers (De Jaeger, R. and Gleria, M., eds.), pp. 61–161. Nova Sciences Publishers, Hauppauge, NY, USA (2007).
2. Varaprath, S., Stutts, D.H. and Kozerski, G.E. A primer on the analytical aspects of silicones at trace levels—challenges and artifacts—a review. Silicon Chem. 3, 79–102 (2006).
3. Warner, N.A., Kozerski, G., Durham, J., Koerner, M., Gerberds, R., Campbell, R. and McNetti, D.A. Positive vs. false detection: A comparison of analytical methods and performance for analysis of cyclic volatile methylsiloxanes (cVMS) in environmental samples from remote regions. Chemosphere 93, 749–756 (2013).
4. Brothers, H.M. Jr, Bovens, E., Bruni, A. et al. A practical gas chromatography flame ionisation detection method for the determination of octamethylcyclotetrasiloxane, decamethylcyclopentasiloxane, and dodecamethylcyclohexasiloxane in silicone emulsions. J. Chromatogr. A 1441, 116–125 (2016).
5. Capela, D., Alves, A., Homem, V. and Santos, L. From the shop to the drain—Volatile methylsiloxanes in cosmetics and personal care products. Environ. Int. 92–93, 50–62 (2016).
6. Capela, D., Homem, V., Alves, A. and Santos, L. Volatile methylsiloxanes in personal care products—Using QuEChERS as a “green” analytical approach. Talanta 155, 94–100 (2016).
7. Wang, R., Moody, R.P., Koniecki, D. and Zhu, J. Low molecular weight cyclic volatile methylsiloxanes in cosmetic products sold in Canada: implication for dermal exposure. Environ. Int. 35, 900–904 (2009).
8. Horii, Y. and Kannan, K. Survey of organosilicone compounds, including cyclic and linear siloxanes, in personal-care and household products. Arch. Environ. Contam. Toxicol. 55, 701–710 (2008).
9. Gouin, T., van Egmund, R., Sparham, C., Hasting, C. and Chowdhury, N. Simulated use and wash-off release of decamethylcyclopentasiloxane used in anti-perspirants. Chemosphere 93, 726–734 (2013).
10. Lewicki, J.P. and Maxwell, R.S. Degradative thermal analysis of engineering silicones. In: Concise Encyclopedia of High Performance Silicones (Tiwari, A., Soucek, M.D., eds.), pp. 191–210. John Wiley & Sons, Hoboken, NJ, USA (2014). https://doi.org/10.1002/9781118384783.ch13.
11. Steimmeyer, R.D. and Becker, M.A. Chromatographic Methods. In: The Analytical Chemistry of Siloxanes (Lee Smith, A., ed.), pp. 255–303. Wiley-Interscience, John Wiley and Sons Inc, New York (1991).
12. Lehotay, S.J., Son, K.A., Kwon, H. et al. Comparison of QuEChERS sample preparation methods for the analysis of pesticide residues in fruits and vegetables. J. Chromatogr. A 1217, 2548–2560 (2010).
Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Shampoo and conditioner lab formulations without added silicone, with added PDMS quaternary ammonium components and with spiked addition of D4, D5, D6 cyclic siloxanes.

Table S2. Composition of lab formulation of shampoo and conditioner with added dimethiconol and amodimethicone