GCMS detection of the presence of EDCs in Black hair care products

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ABSTRACT

Personal care products have been shown to contain endocrine-disrupting chemicals (EDCs), which can pose as hormones and interfere with the endocrine system. This interference can lead to obesity, cancer, and reproductive/fertility issues. In previous studies, researchers have suggested that certain hair care products (e.g., hair relaxers, hair oils, and hair moisturizers) have higher concentrations of EDCs than others, and these specific hair care products are more commonly used by Black women. Therefore, the use of these products may be presenting an increased health risk to Black women. We used gas chromatography-mass spectrometry (GC/MS) to test 10 different products in 3 different categories used by black women: moisturizers, hair oils, and relaxers. There were 3 different categories of chemicals that we aimed to test: cyclosiloxanes, parabens, and phthalates. Due to study limitations, cyclosiloxanes were the only EDCs detected. All three of the hair product categories detected an EDC. Haircare products promoted toward Black women contained EDCs and were not always listed on ingredient labels.

INTRODUCTION

Many people believe that hair care products are only impacting the hair on the outside; however, there are harmful EDCs present within the products that cause negative health effects on the body. Various studies show there are in fact EDCs present as well as explain the negative impact they have on the endocrine system. If the public is informed on what exactly is found within the hair care products and what the effects of exposure to those chemicals could do, more precautions will be taken both by the companies as well as consumers. This study aims to bring light to this information to the Black community by measuring what EDCs are found in hair care products that are used more frequently by African American women. This community is being targeted in this study because the hair products being tested are used more frequently by African American women and there are higher rates of health disparities.

Endocrine System

The endocrine system is a series of glands that produce and secrete hormones that the body uses for a wide range of functions. These control many different bodily functions, including respiration, metabolism, reproduction, sensory perception, movement, sexual development, and growth. The system uses homeostasis, which is a system that regulates the internal conditions of the body. In Figure 1. It shows the organs and glands in the endocrine system that the system uses to send hormones through the body to produce a message to the body. The endocrine system maintains homeostasis by regulating bodily functions, for instance, body temperature may be regulated by sweating or shivering. The endocrine system uses hormones to send messages through the body to tell the body to react accordingly. Figure 1. shows the organs and glands in the endocrine system that produce hormones that the endocrine system uses to send messages through the body to produce an action for the body to do . For example, the testicles and the ovaries produce hormones such as testosterone and estrogen. Those two organs are a part of the endocrine system. These hormones are produced for the reproduction process. Some people have endocrine disorders due to an underproduction of a hormone (e.g., insulin in diabetes). The endocrine system uses the pancreas organ to produce the insulin hormone. Other people may have an overproduction of a hormone (e.g. thyroid-stimulating hormone -TSH - in Graves

disease). The endocrine system's standard functions can be disturbed by chemicals outside the body (i.e. exogenous chemicals) classified as endocrine-disrupting chemicals (EDCs).(1)

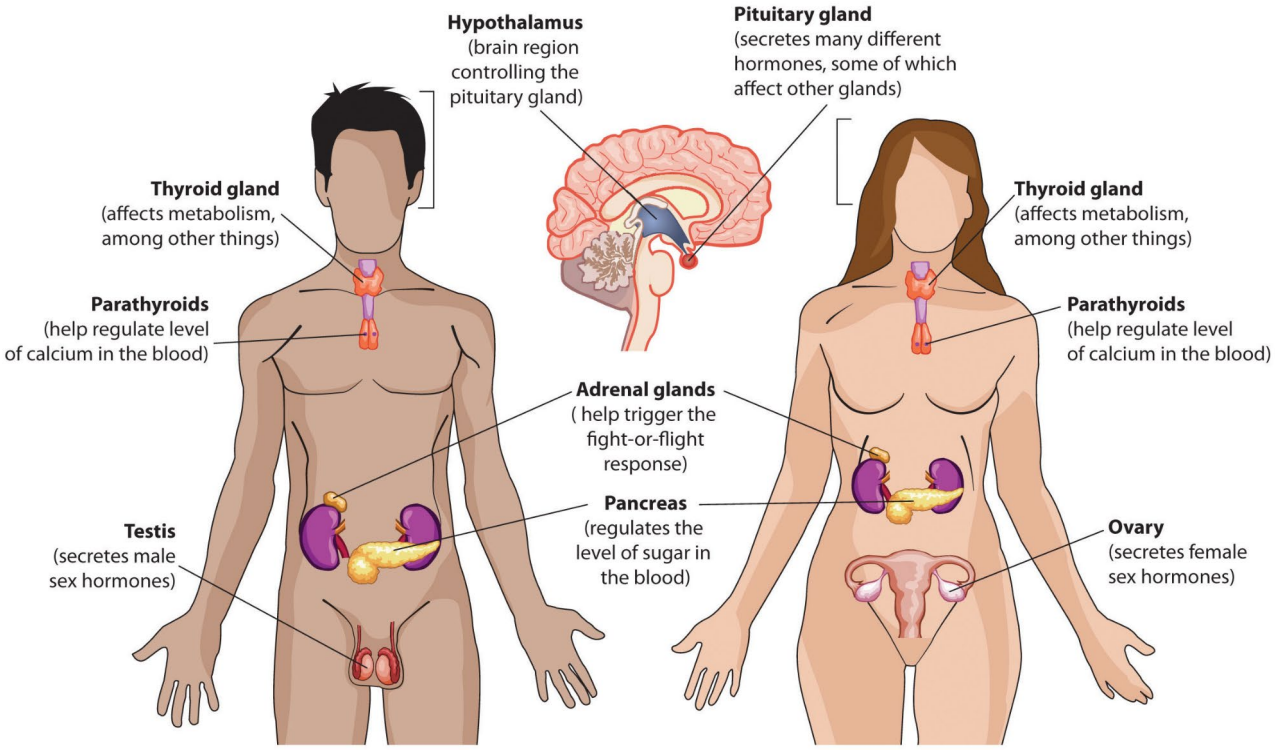


Figure 1. The endocrine system *(2)*

Endocrine-Disrupting Chemicals (EDCs)

EDCs are "an exogenous chemical or mixture of chemicals, that interferes with any aspect of hormone action" (3). There are a couple of common EDCs, such as Bisphenol A (BPA) and parabens. Some banned EDCs are polychlorinated biphenyls (PCBs) and lead. Some EDCs have been banned, and many companies may advertise their products as "free" from others, such as BPA-free and paraben-free. Exposure to EDCs is nearly impossible to avoid just because many everyday consumer products contain these chemicals. They can be unavoidable because the ingredient labels can be inaccurate. Many chemicals that were found within the products were not distinctly listed on the label of ingredients (4). Exposure to EDCs may lead to negative health effects, such as obesity, diabetes, reproductive issues, and breast cancer. A review by the Endocrine Society comprehensively describes evidence from both animal models and epidemiological studies connecting EDCs to an increased risk of these diseases. Specifically,

Gore et al. summarized research primarily from 2010 to 2015 focused on exposure to BPA, phthalates, and parabens as well as others (5).

The effects of phthalates are of particular interest to our proposed study, as these chemicals have been found in hair care products(6). Exposure to phthalates has been related to negative effects on uterine structure/ function, which was shown in a previous study (7). Parabens have also been studied, and in the studies that have been done, it has been concluded that EDCs have been linked to health issues such as menarche and breast cancer. Studies have shown that EDCs not only affect people once they are adults but using products that contain EDCs at a younger age can cause these health problems in the future. (6)

Methods for analyzing EDCs

Chromatographic studies of EDCs in personal care products (PCPs) from the past 10 years have been reviewed; 70 of those studies used chromatography and 59 used liquid chromatography. Of the review studies over 50% tested for phthalates and parabens and 9% tested organosiloxane. There are often three steps in analyzing the PCPs which are the following: pretreatment, extraction and identifying the compound. In most cases prior to chromatographic analysis PCP samples typically have a pretreatment of methods such as mixing and extracting. mixing can include shaking, sonicating, centrifuging, and vortexing. extracting can include any of the techniques shown Figure 2. (8)

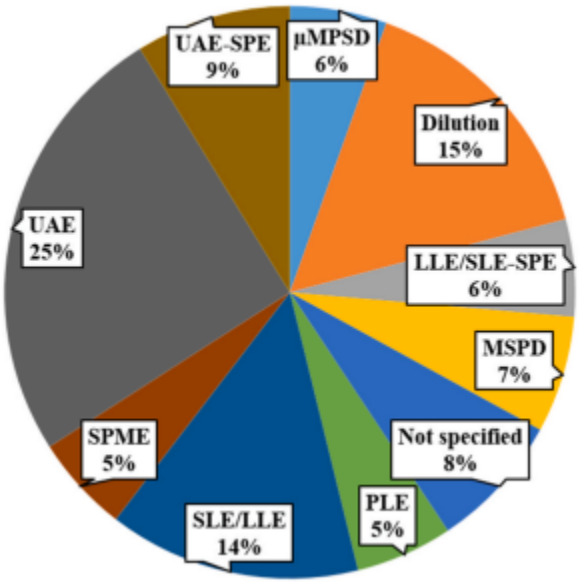


Figure 2. Most used extraction techniques in a reviewed study (8).

When using GC, derivatization may be required in order to allow peak separation. In the study compounds tested for polar and needed to be delivered ties to perform a good chromatographic column. Of the compounds tested 34% were derivatized. In some cases the samples were also concentrated and or filtered as part of their pre-treatment. The common products that were tested range from sunscreen care products, sunscreen cosmetics, hair care products, body care products, feminine products, dental care products, makeup products, and baby care products. The common EDCs measured are the following: preservatives, parabens, phthalates, Antiseptics/disinfectants,UV-filters, musk fragrances, bisphenols, alkylphenols, siloxanes, and other chemicals, The company can be classifying certain chemicals under the fragrance term. The FDA essentially allows companies to keep the ingredients to their products classified so that the formula does not get spread and keep their products unique. (6)

EDCs have been detected in a wide variety of consumer products that did not disclose their presence on the label. Helm et al. tested 18 products under the following six different categories: hot oil treatments, anti-frizz, leave-in conditioner, root stimulator, hair lotion, and relaxer. The products were selected along two guidelines which are why 12 of the products were chosen because they are the most frequently used and the other 6 were chosen because they are used by black women the most. The 66 chemicals that were chosen to be analyzed based on the relevance they had to disruption to the endocrine system and asthma, as well as the probability that it would even be found in the product (6). Theproducts did not list 84% of the chemicals detected in the products on the labels. The concentration of the chemicals in the products being tested were low, soonly 8% of the time the chemicals that were present got listed on the label. Still, a product with even more of a higher concentration of the chemicals being tested, were listed around 50% of the time. This study tested three different chemicals that stood out to me based on the products presenting them. The types of hair care products that I am using contain the highest concentration levels of the chemicals I am studying, which are the following: Decamethylcyclopentasiloxane which is used for conditioning and spreadability in hair products. In this study, 67% of products detected at least one of the three target cyclosiloxanes. Methylparaben acts as a preservative in hair products, and 72% of products detected this chemical. Lastly, Diethyl phthalate, which is used as a solvent in fragrances within hair products, are detected in 78% of the hair products that were being tested. The following structures of chemicals are shown in figure 3.

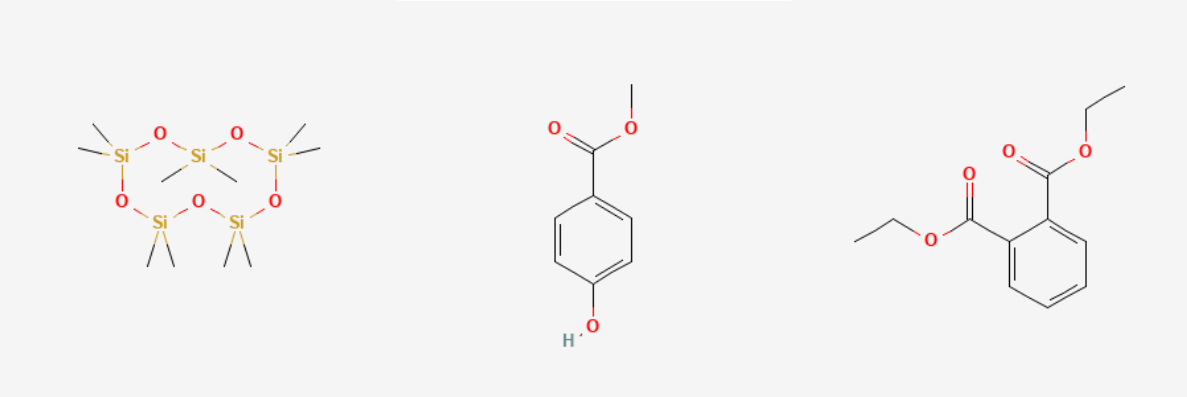


Figure 3. Structure of Decamethylcyclopentasiloxane, Methylparaben, and Diethyl phthalate.

GCMS

The instrument separates chemical mixtures (the GC component) and identifies the components at a molecular level (the MS component). The GC works on the principle that a mixture will separate into individual substances when heated. The heated gases are carried through a column with a gas (such as helium). As the separated substances emerge from the column opening, they flow into the MS. Mass spectrometry identifies compounds by the mass of the analyte molecule. A library of known mass spectra, covering several thousand compounds, is stored on a computer.

EXPERIMENTAL

Chemicals and Equipment

HPLC-grade solvents, dichloromethane (99.8%), and methanol (99.9%), were purchased from Acros Organics. Supelco Discovery ® DSC−NH_2 SPE Tubes (100 mg, volume: 1mL) for solid-phase extraction were purchased from Sigma-Aldrich. GCMS analysis was performed using a GCMS-QP2010 Shimadzu Gas Chromatograph Mass Spectrometer consisting of an AOC-5000 Auto-Injector and an SH RXI-5MS column (25 mm thickness, 30 m length, and 25 mm diameter), and operated using GCMSsolution Software. Black hair care products were purchased online from Amazon, in-store at a local beauty supply store, and also in-store at a

local Target. The ten products included four moisturizers, three hair oils, and three relaxers (two relaxer kits and one relaxer cream) as shown in table 1.

Selection of Hair Care Products

A list of potential Black hair care products was generated through products found in stores as well as through an Internet search for “hair lotions and or blue moisturizers promoted towards black women " and “oils promoted towards black women ". There were two search results that we utilized, both were lists of products from the website: stylecraze.com the lists included the following, “11 Best Hair Moisturizers For African American Hair” and “13 Best Hair Oils For Black And Kinky Hair". In our location: Pittsburgh, PA there is a sky’s the limit: a beauty supply store in a predominantly Black neighborhood that is advertised toward black women as you can see in figure 4. A nearby Target that sells hair products that are promoted towards Black women in a section that is labeled “textured hair care” as seen in figure 5, is another store where we saw potential products. The products chosen are listed in the chart below. In order to decide which products were going to be used it was solely based on the ingredient labels on the products. The hair products were classified into 3 different categories under the following: containing the EDC (i.e. explicitly listed), not containing the EDC (i.e. “paraben-free” noted on the product), or unknown (i.e. did not list nor did it say it was free of the EDC). So after dividing the categories up, gathering a variety of products was prioritized which is why in each type of product (i.e. moisturizer, hair oil, or relaxer) there is a product that either contains, is free of, or does not list a specific chemical. In trying to select a variety of products, it came to the conclusion that none of the products listed diethyl phthalate directly. Therefore, we do not have that category but there were products that listed “phthalate-free” on the product. We also concluded that decamethylcyclopentasiloxane was not listed on any of the products as “free of”, but it did directly list that the products contained decamethylcyclopentasiloxane. The last chemical methylparaben was both listed “free of” and directly listed that the product contains it. The products selected have not been tested in the 2018 paper that I referenced.

Figure 4. The advertisement for Black hair care products in target

Figure 5. The advertisement for Black hair care products at a local beauty supply store

Table 1. Black hair care products tested

Product Category	Product name	Diethyl phthalate	Methylparaben	Decamethyl-cyclopentasiloxane
Moisturizer	Thank God It's Natural Butter Cream Daily Moisturizer[a]			
	Natural FORMULA Natural Formula Hair Moisturizer Go Curly for Building Curls[a]			
	Cantu curling cream[b]			
	Shea moisture leave-in conditioner[c]			
Hair oil	African Pride Moisture Miracle 5 Essential Oils[a]			
	Palmers cocoa butter moisturizing hair oil[c]			
	Doo Gro mega thick formula hair oil[c]			
Relaxer	Luster's Pink conditioning no lye relaxer[b]			
	Creme of nature advanced straightening with exotic shine[b]			
	Soft Sheen carson optimum smooth relaxer system[b]			

Key: black = listed; white = free of the chemical; gray = not listed in ingredients, but not identified as "free" of the chemical

[a] Amazon.com; [b] Sky's the Limit local retailer; [c] Target

Preparation of samples

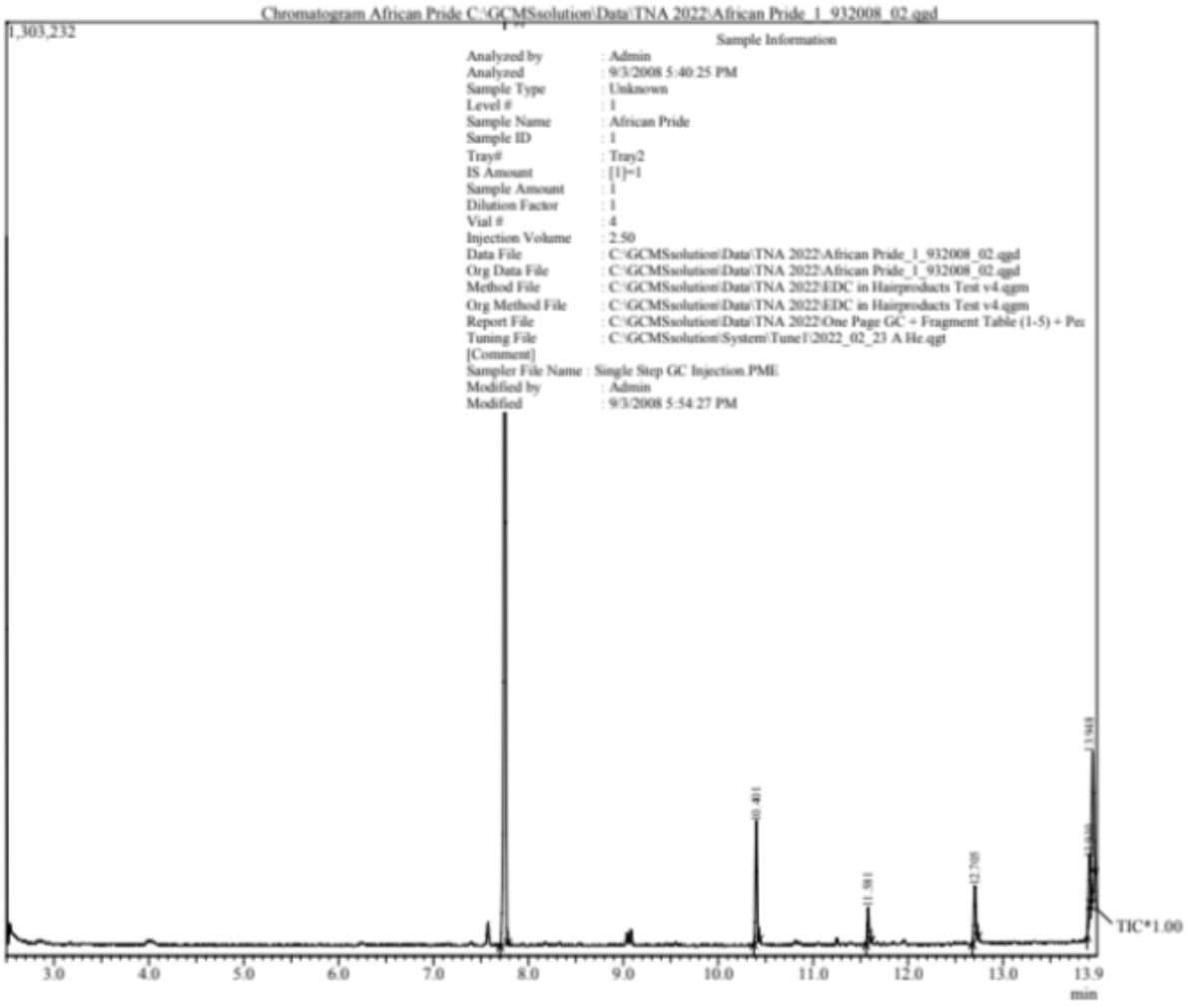
An aliquot (0.25 mL) of each hair care product was dissolved in 50 mL of 3:1 dichloromethane:methanol for 3-5 mins with mechanical stirring. Six milliliters of each hair product solution was passed through a SPE cartridge and 0.100 mL was diluted with 1.5 mL of methanol for GCMS analysis.

GCMS Analysis

The hair care product samples were run at a flow rate of 6.9 mL/min for 14 minutes with a split injection ratio of 5:1. The initial oven temperature of 40 °C was increased at a rate of 20 °C/min for 10 mins to reach a final temperature of 240 °C and held for 4 min. The cut time of the detector to begin recording was 2.5 min to eliminate the methanol peak. The highest peaks between 45-450 m/z were automatically picked by the software and their mass fragment tables compiled and compared to a database of known substances.

RESULTS

A representative chromatogram is shown in Figure 6 and a mass spectrum for decamethylpentacyclosiloxane is given in Figure 7. A variety of cyclosiloxanes were detected in the study, including: hexamethylcyclotrisiloxane (4.7 min),-octamethylcyclotetrasiloxane (6.4 min), decamethylcyclopentasiloxane (7.5 min), dodecamethylcyclohexasiloxane (9.04 min). Table 2 summarizes which hair products contained a chemical. Methylparaben is excluded from this table because we were unable to prepare samples for this specific analysis due to supply chain issues. Argan oil relaxer kit conditioner was also excluded because we were unable to collect a sufficient volume of sample following solid phase extraction. The dissolved product did not easily pass through the SPE cartridge and we did not have a set-up that could apply pressure to force the mixture through.



Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height	Height%	A/H	Mark	Name
1	7.752	7.703	7.785	1655155	66.83	1287604	69.30	1.29		Cyclopentasiloxane, decamethyl-
2	10.401	10.375	10.433	215518	8.70	169031	9.10	1.28		Dodecanoic acid, methyl ester
3	11.581	11.559	11.612	64686	2.61	49473	2.66	1.31		Methyl tetradecanoate
4	12.705	12.679	12.738	111884	4.52	76909	4.14	1.45		Hexadecanoic acid, methyl ester
5	13.910	13.887	13.928	142649	5.76	83165	4.48	1.72		Methyl 10-trans,12-cis-octadecadienoate
6	13.948	13.928	13.968	286689	11.58	191950	10.33	1.49	V	10-Octadecenoic acid, methyl ester
				2476581	100.00	1858132	100.00			

Figure 6. Gas chromatogram of African Pride Moisture Miracle 5 Essential Oil.

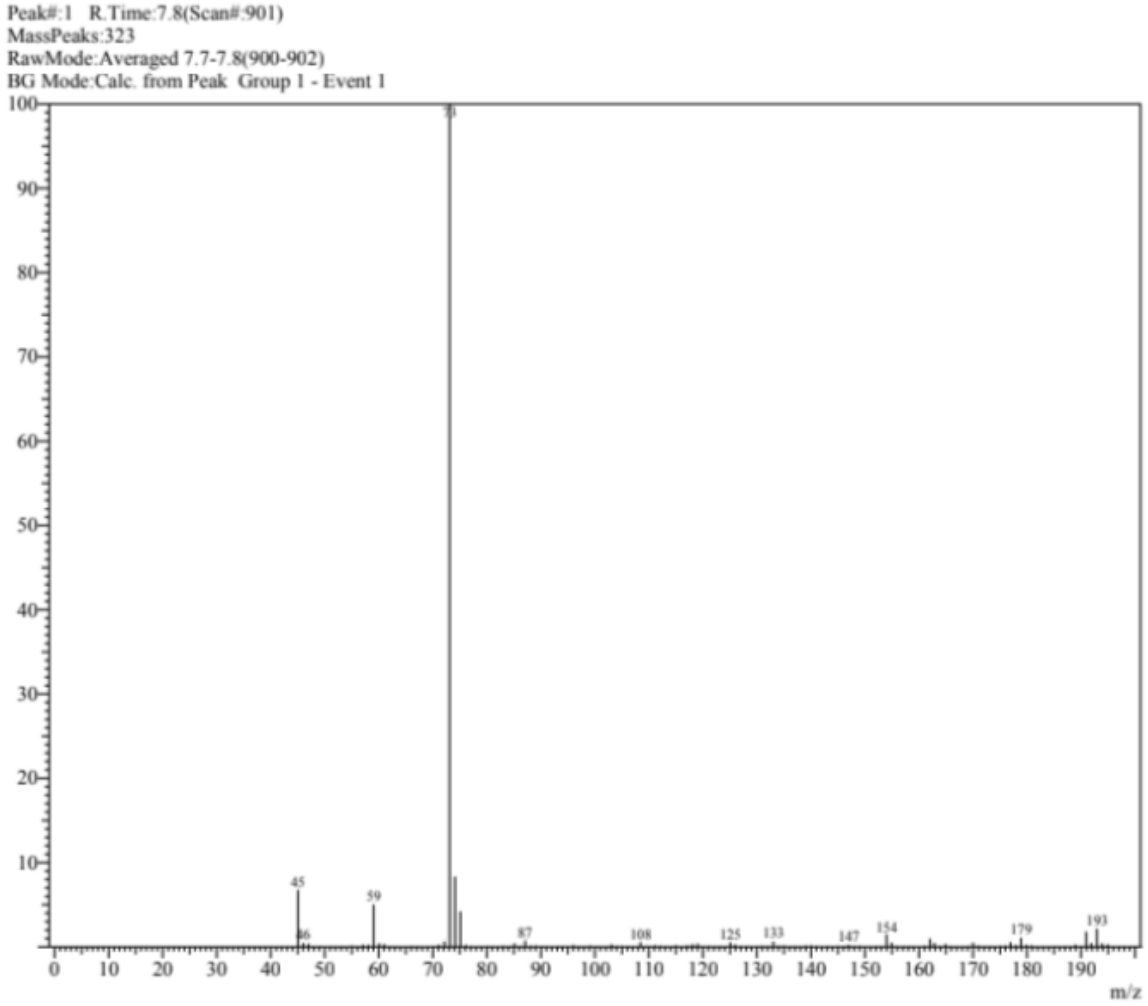


Figure 7. Mass spectrum of decamethylcyclopentasiloxane

Table 2. The hair care products table of rather or not a chemical was detected or not.

Product Category	Product name	diethyl phthalate	cyclosiloxanes
Moisturizer	Thank God It's Natural Butter Cream Daily Moisturizer[a]		detected
	Natural FORMULA Natural Formula Hair Moisturizer Go Curly for Building Curls[a]		
	cantu COCONUT CURLING CREAM[b]		
	Shea Moisture Leave in Conditioner with Jamaican Black Castor Oil for Hair Growth, Strengthen & Restore, Vitamin E[c]		detected
Hair oil	African Pride Moisture Miracle 5 Essential Oils[a]		detected
	Palmer's Cocoa Butter Formula Moisturizing Hair Oil[c]		
	Doo Gro Mega Thick Hair Growth Oil[c]		
Relaxer	Pink Conditioning No-Lye Relaxer Retouch Kit[b]		detected
	Argan Oil Relaxer provides advanced straightening performance and Exotic Shine™[b]		
	Soft Sheen carson optimum smooth relaxer system[b]		detected

DISCUSSION

In this study we aimed to detect the presence of three EDCs in hair care products: diethyl phthalate, methylparaben, and decamethylcyclopentasiloxane. There was one EDC detected, one EDC undetected, and one EDC unable to be tested.

Cyclosiloxanes

Four different types of cyclosiloxanes were detected in five different products and they were in all three of the diffrent hair product type categories. There were two moisturizers that had the cyclosiloxanes detected.In the TGIN there was octamethylcyclotetrasiloxane and decamethylcyclopentasiloxane, and in the Shea moisture there was dodecamethylcyclohexasiloxane. There was one hair oil that detected cyclosiloxanes, which was African Pride. This hair oil contained both decamethylcyclopentasiloxane and dodecamethylcyclohexasiloxane. There were two relaxers that had cyclosiloxanes detected. In the Luster relaxer there was octamethylcyclotetrasiloxane, decamethylcyclopentasiloxane, and hexamethylcyclotrisiloxane. 60% of the products that detected cyclosiloxane did not have that listed on their product labels. In the case of shea moisture it was detected in a lower quantity so that could be why they did not include it on the label. In the Soft Sheen carson optimum smooth relaxer system, there was dodecamethylcyclohexasiloxane. There were 50% of products that detected one of four cyclosiloxanes. In previous studies there were 67% of of products that detected at least one of the three target cyclosiloxanes (2018).

Diethyl phthalate

There was no diethyl phthalate detected in any of the products, which was likely because the boiling point is 295 ℃ and the method we used only required the oven to be set to 240 ℃. Therefore, the oven was not set to a high enough temperature actually hot enough to reach the boiling point. Although the results confirm the ingredient labels, the detection of this chemical was expected because in previous studies if was shown to be found in many products.

Methylparaben

We were unable to include the testing of methylparaben because the solvent that was going to be used in the procedure to derivatize the methylparaben to lower the boiling point for the gas chromatography, was back-ordered due to supply chain issues during the COVID-19 pandemic. This chemical was the one we were most interested in detecting, because we had

products that fit each label criteria, (e.g., listed, "free", and not listed). I expected it to be detected in many of the products because in previous studies it has been shown to be found in 72% of products tested, not only that but it is a commonly known EDC. (6)

Limitations and suggestions

Not all products got tested because when making the samples the solution was transferring to slow through the solid phase extraction cartridges, and ended up evaporating before fully passing through. In the future allowing a longer run time through the GCMS for further analyzing because depending on the affinity of the chemical the chemicals could have stuck to the column longer than 14 minutes. I would also recommend testing for concentration rather than just the detection so that the amount presented could be determined.

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