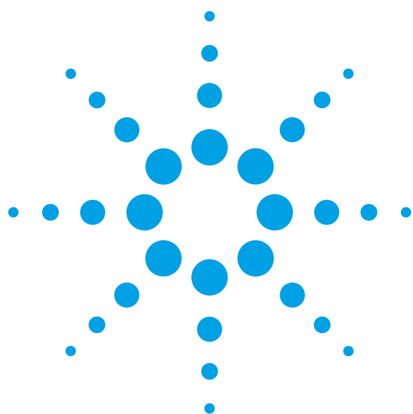

Determination of Parabens in Body Wash using Solid-Supported Liquid-Liquid Extraction

Application Note

Consumer Products

Authors

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Abstract

Parabens are compounds often added to cosmetics such as lotions, deodorants, and body washes as preservatives. Although they are useful as preservatives, studies have linked these compounds to several adverse side effects such as cancer, infertility, and miscarriage. The extraction of four parabens (methyl paraben, ethyl paraben, propyl paraben, and butyl paraben) from an infant shampoo/body wash was studied using Agilent Chem Elut 5 mL unbuffered solid-supported liquid-liquid extraction (SLE) cartridges. An Agilent 1200 HPLC system with diode array detection was used for the separation and determination of the extracted parabens. The recoveries of the parabens ranged from 82% to 101% when extracted using the SLE method.

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Introduction

Parabens are a family of compounds that are often used to stop the growth of fungi, bacteria, and other microbes in cosmetic products. Parabens are the most widely used preservatives in personal care products [1]. As such, they have been under scientific scrutiny because they have been linked to reproductive issues by some studies [2]. There has been much research focused on parabens because they are so widely used. Studies have shown that parabens can mimic estrogen, which can trigger the development of cancerous cells. Parabens have been linked to breast cancer because they have been found amongst breast cancer tumor cells [3]. Since parabens mimic estrogen, they are also thought to mimic its adverse effects. Due to their presence in many skin cosmetics, exposure to parabens is typically high.

This application note used solid-supported liquid-liquid extraction (SLE) for the extraction of parabens from a shampoo/body wash. This was followed by high performance liquid chromatography (HPLC) for quantitative analysis of the parabens extracted by the SLE method. Additional samples of the shampoo/body wash were extracted using a liquid-liquid extraction (LLE) so that the two methods could be compared.

In SLE, a high purity, finely divided, inert, diatomaceous earth sorbent is used to aid the extraction of the analyte from an aqueous solution into an organic solvent. The aqueous solution containing the analyte is passed through the cartridge and the aqueous phase is adsorbed onto the diatomaceous earth. Once the solution has been adsorbed onto the sorbent, an immiscible organic solvent is used to extract and elute the analyte off the cartridge [4]. Because the aqueous solution is spread over the sorbent in a very thin layer, the two solvents are in intimate contact and the analyte can be extracted into the organic solvent without the shaking necessary in LLE. This helps to avoid the problem of emulsion formation that is common in LLE. SLE cartridges typically incorporate a phase separation filter at the outlet to prevent mixing which results in elution of the aqueous phase along with the organic solvent. Chem Elut SLE is available in several formats, and can be purchased in prepacked cartridges or by bulk.

Experimental

Ultrapure water was delivered using a Millipore Synergy UV purification system. Acetonitrile (Burdick and Jackson, Muskegan, MI), methanol (Fisher Scientific, Fairlawn, NJ), acetone and ethyl acetate (Pharmco, Brookfield, CT) were HPLC grade. Methyl paraben, ethyl paraben, propyl paraben, and butyl paraben were purchased from Sigma-Aldrich Corp. The stock solution, including each of the parabens at a concentration of approximately 10 mg/mL, was prepared in methanol. Standard solutions were prepared in methanol by dilution of the stock solutions. Standards were prepared at nominal concentrations of 1000, 500, 100, 50, 25, 10, and 1 µg/mL.

Extractions

The prepared stock solution was used to spike the body wash to determine the recoveries. The method used was adapted from a published SLE method developed for measuring pesticide residues in honey [5,6]. The sample was prepared as described in Figure 1 prior to performing the extractions. The entire prepared sample was poured onto the SLE cartridge for the extraction step. Twenty four samples were prepared as described in Figure 1. Twelve were spiked at 175 µg/mL and 12 were spiked at 20 µg/mL so that recoveries could be calculated at both high and low paraben levels. Of the 12 spiked at each level, six were extracted using the SLE method described in Figure 2, and the other six were extracted using the LLE method described in Figure 3 so the two methods could be compared. The SLE cartridges used were Chem Elut, 5 mL unbuffered.

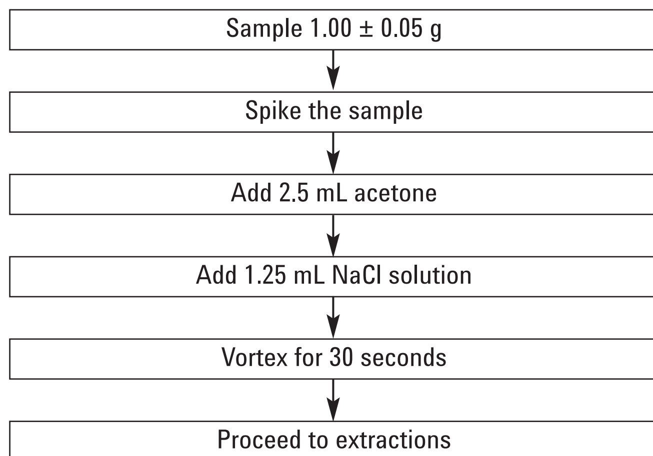

Figure 1. Preparation of sample prior to extraction by SLE or LLE.

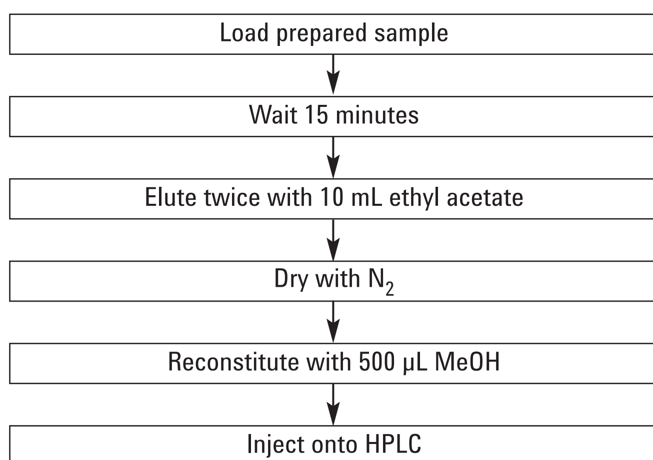

Figure 2. Procedure used to extract parabens from shampoo/body wash using SLE.

HPLC

The analysis was performed on an Agilent 1200 Infinity Series with a binary pump, autosampler, inline degasser, and an 80 Hz Diode Array Detector. The detector flow cell chosen for this study was a micro flow cell with a 2 µL volume. ChemStation for LC 3D Systems, Rev. B.03.01, was used for data collection and analysis.

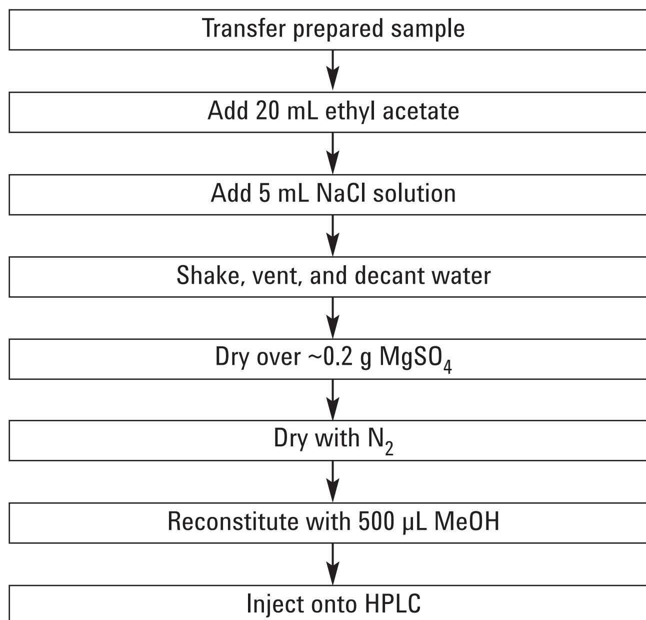

Figure 3. Procedure used to extract parabens from the shampoo/body wash using LLE in a separating funnel.

The analytical column was an Agilent ZORBAX Eclipse Plus C18. The run time was 9 minutes with a re-equilibration time of 2 minutes.

Conditions

Column:	Agilent ZORBAX Eclipse Plus C18, 4.6 x 150 mm, 5 µm (p/n 959993-902)	
Sample prep:	Agilent Chem Elut, 5 mL (p/n 12198006)	
Eluent:	A, 90% Water: 10% acetonitrile; B, acetonitrile	
Injection volume:	1.7 µL	
Flow rate:	2.00 mL/min	
Gradient:	Time (min)	% B
	0.00	30
	4.00	65
	5.00	70
Response time:	0.02 s	
Detection:	230 nm	

Results and Discussion

The resulting calibration curves for the four parabens are given in Figure 4. Table 1 shows the linear regression results for the calibration curves.

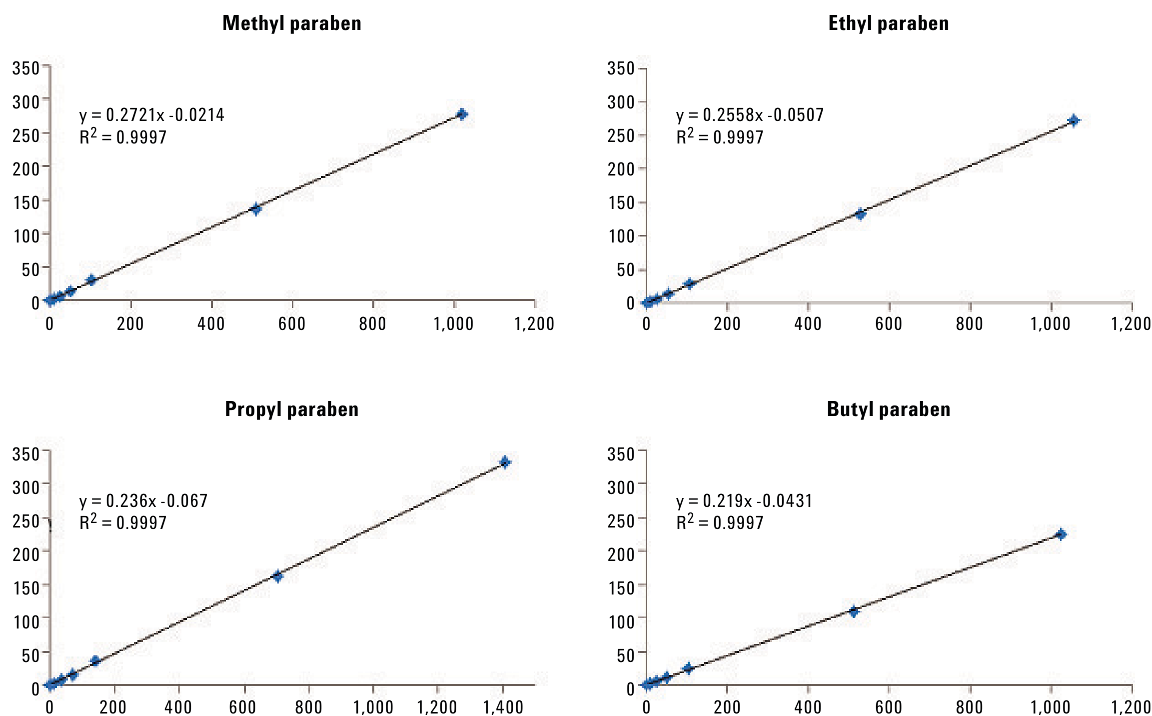

Figure 4. Calibration curves of four parabens.

Table 1. Linear regression results for calibration curves of four parabens.

Compound	Least squares line of best fit	R^2
Methyl paraben	y = 0.2721x – 0.0214	0.9997
Ethyl paraben	y = 0.2558x + 0.0507	0.9997
Propyl paraben	y = 0.2360x + 0.0670	0.9997
Butyl paraben	y = 0.2190x + 0.0431	0.9997

Chromatograms of the shampoo/body wash (not spiked) after extraction by SLE and LLE are shown in Figure 5. This sample was chosen because the label stated it did not contain any parabens. Chromatograms of the spiked samples after extraction by SLE and LLE are shown in Figure 6. The chromatograms indicate that in the area of the chromatogram that the parabens elute, the sample extracted by SLE (shown in Figures 5A and 6A) was much cleaner and none of the peaks that were present in the unspiked sample coeluted with the analytes of interest. The sample that was extracted by LLE (shown in Figures 5B and 6B) had interferences extracted from the matrix, along with the analytes of interest. These interference peaks made it difficult to accurately quantify the peak for methyl paraben and butyl paraben.

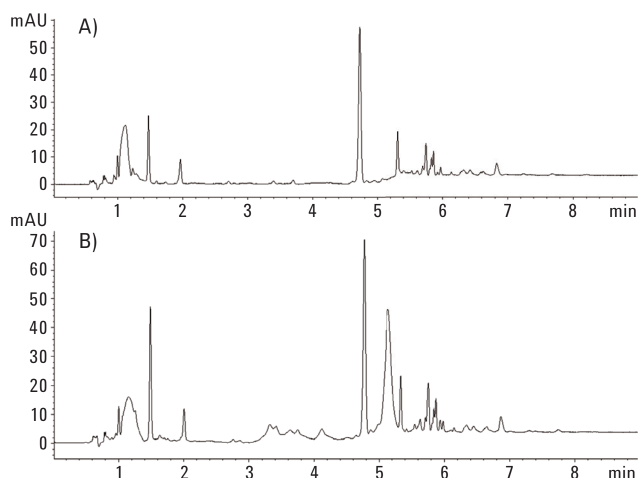

Figure 5. Chromatograms of infant shampoo/body wash (not spiked) after A) SLE, B) LLE.

Table 2 shows the calculated recoveries for the four parabens after extraction from the shampoo/body wash by SLE and LLE. Note that although the low concentration samples were spiked at 20 µg/mL, the injected concentrations were 40 µg/mL since the extracted samples were dried and reconstituted in half the original volume. Both techniques gave reasonable values for percent recovery, but the values obtained when the extraction was done by SLE were better. In general, the percent recovery by SLE was higher, and in all cases the standard deviations were lower

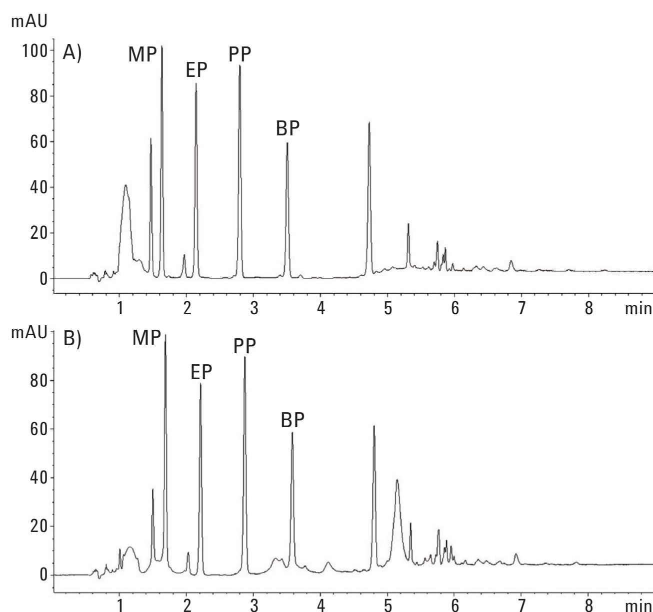

Figure 6. Chromatograms of spiked shampoo/body wash after A) SLE, B) LLE.

Table 2. Calculated percent recoveries for the extraction of four phthalates from infant shampoo/body wash using SLE and LLE.

	% Recovery (LLE)				% Recovery (SLE)			
	Spiked at 20 µg/mL		Spiked at 175 µg/mL		Spiked at 20 µg/mL		Spiked at 175 µg/mL	
	avg	std dev	avg	std dev	avg	std dev	avg	std dev
Methyl paraben	94.32	17.82	79.15	2.53	96.87	2.33	100.29	1.33
Ethyl paraben	83.14	7.63	81.80	2.95	87.78	3.68	101.04	0.78
Propyl paraben	81.95	6.16	83.93	3.08	82.53	3.94	99.87	1.42
Butyl paraben	97.36	26.54	82.94	4.86	84.26	3.79	99.41	1.21

Conclusions

The results show that Agilent Chem Elut SLE cartridges offer an effective method for the extraction of parabens from a shampoo/body wash matrix. The impurities that were extracted from the matrix together with the parabens were minimal and did not interfere with the quantitation of the analytes. When compared to LLE, the results produced by SLE were superior.

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Printed in the USA
July 11, 2013
5991-2735EN

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