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Analysis of consumer cosmetic products for phthalate esters

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Synopsis

A rapid and sensitive reverse-phase HPLC method with UV detection was developed for the quantitation of dimethyl phthalate (DMP), diethyl phthalate (DEP), butyl benzyl phthalate (BBP), dibutyl phthalate (DBP), and di(2-ethylhexyl) phthalate (DEHP) in cosmetic preparations. Average recoveries of the phthalate esters were better than 90%. In a survey of 48 consumer cosmetic products, including hair care products, deodorants, lotions and creams, nail products, fragrances, and body washes, most products were found to contain at least one phthalate ester. DEP was detected most frequently at concentrations up to 38,663 ppm. DBP was found in fewer products, but at levels up to 59,815 ppm. Based on the available exposure and toxicity data, the FDA has concluded that there is insufficient data to conclude that a human health hazard exists from exposure to phthalate esters from cosmetic products.

INTRODUCTION

Phthalate esters are present in many consumer products, primarily to impart flexibility to rigid polymers such as polyvinyl chloride. They are used in the production of products such as food wrappings, medical devices (e.g., blood bags), children's toys, wood finishes, paints, upholstery, and plastic products, and are subject to a variety of regulatory requirements. As a result of their extensive use, phthalate esters have been found in the environment (1–6), foods (7–9), food supplements (10), medical products (11,12), medical devices (13), plastic materials (14), and cosmetics (15). In cosmetic products, phthalate esters are used as solvents for fragrances, as suspension agents for solids in aerosols, as lubricants for aerosol valves, and as antifoaming agents, skin emollients, and plasticizers in nail polishes and fingernail elongators.

Investigations into the levels of phthalate ester metabolites in human urine have shown that exposure to DEP, BBP, DBP, and DEHP from all sources is highly variable from person to person and between different demographic groups (16–19). In one study, women of child-bearing age (20–40 years) were found to have significantly higher levels of monobutyl phthalate, the metabolite of DBP, in their urine than other age/gender groups (16). Several phthalate metabolites have also been found in human breast milk

(20). Estimates of daily human exposure to DEP, based on ester metabolite concentrations in urine, range from undetectable to 320 mg/kg/day (21).

Phthalate esters, are of potential concern because of reproductive and other toxic effects reported for phthalate esters in animal models. Tests with rats have shown that some phthalate esters can damage the male reproductive system of offspring and cause other developmental abnormalities (22–36).

The relevance of human exposure to phthalate esters is difficult to assess because the effects observed in animals resulted from exposure to relatively high doses (on the order of 100 to 1000 mg/kg/day), and more importantly, rats may metabolize phthalate esters differently than humans (37,38). While one study using a comet assay suggested that the urinary metabolite of DEP at levels currently found in the environment might cause human sperm DNA damage (39), another study showed primates were less sensitive to phthalate exposure than rodents (40). Further, because of their ubiquitous use, it is difficult to pinpoint any specific source of phthalate ester uses as being potentially responsible for any observed effects.

In July 2002, the Environmental Working Group (EWG), a coalition of environmental and public health organizations, reported on the analysis of 72 cosmetic products for the phthalate esters DMP, DEP, BBP, DBP, and DEHP (15). According to the report, phthalate esters were found in 52 of the 72 products tested, at levels ranging from less than 50 parts per million to nearly three percent. Also, according to the report, none of the 52 products listed the phthalate esters as ingredients on the product labels. Based on the safety concerns raised by the EWG and other published data, the FDA initiated a project to determine consumer exposure to phthalate esters from cosmetic products (41). The present study was undertaken to develop and validate an analytical method for phthalate esters in cosmetic products, to verify the levels reported in cosmetics by the EWG, and to collect additional data on phthalate ester levels in other types of cosmetic products.

A large variety of analytical methods have been published for the analysis of consumer products, biological materials, and environmental samples for phthalate esters. Methods based on gas chromatography with either flame ionization or electron capture detection have been described for food simulants (42), water (43), plasma (44) and edible oils (45). High-performance liquid chromatography (HPLC) with UV detection has been applied to water (46), IV drug solutions (47), blood plasma (48), and water (49). More recently reported methods have utilized mass spectrometry for the detection of phthalate esters. Methods applying gas chromatography coupled to a mass spectrometer have been described for the analysis of water (50–53), saliva (54), and plastic and PVC materials (55,56). HPLC and mass spectrometry have been used for the analysis of urine (57–59), human milk (20), and IV drug solutions (60). Very little has been published on analytical methods for the determination of phthalate esters in cosmetic products. Two older methods, based on simple gas chromatography, have been described for cosmetic products (61,62), but little data has been provided. The present study describes a method for the determination of phthalate esters in a variety of different cosmetic products. The method utilized a Celite column extraction method originally developed in our laboratory for the analysis of phenol, resorcinol, salicylic acid, and α-hydroxy acids in cosmetic products and salon preparations (63). The method was validated for the determination of phthalate esters in several different types of cosmetic products.

EXPERIMENTAL

REAGENTS AND MATERIALS

The following reagents and materials were used: Hexane was purchased from Burdick & Jackson (Muskegon, Michigan). Acetonitrile and methanol were purchased from T. J. Baker (Phillipsburg, New Jersey). 2-Propanol was purchased from Fisher Scientific (Fairlawn, New Jersey). All solvents were HPLC grade. Phthalate esters DMP (99%), DEP (99.5%), and BBP (98%) were purchased from Sigma Aldrich (Milwaukee, Wisconsin). DBP (≥98%) and DEHP (99.5%) were purchased from Sigma Aldrich (St. Louis, Missouri). De-ionized water was prepared with a Milli-Q purification system from Millipore (Billerica, Massachusetts). Celite 545 was purchased from Fisher Scientific (Fairlawn, New Jersey). The extraction tubes and filter disks were obtained from Supelco (Bellefonte, Pennsylvania).

PHTHALATE ESTER CALIBRATION STANDARDS

A primary standard solution of a mixture of the five phthalate esters (~ mg/ml each) was prepared by adding approximately 100 mg of each to a 100-ml amber volumetric flask and diluting to the mark with hexane. Because of the wide range of possible concentrations in cosmetic products, three sets of working standards were prepared. One set was prepared at approximately 0.001, 0.003, 0.006, and 0.01 mg/ml by appropriate serial dilution of the stock solution. Similarly, a second set was prepared at approximately 0.01, 0.03, 0.06, and 0.1 mg/ml, and a third set for BBP, DBP, and DEHP only was prepared at 0.10, 0.30, 0.60, and 1.00 mg/ml. HPLC peak areas were determined based on duplicate injections of 20 µl, and a calibration curve was obtained by plotting peak area versus standard concentration.

SAMPLE EXTRACTION

To avoid contamination by environmental sources of phthalate esters, all glassware was thoroughly cleaned and rinsed with water and ethanol before use, and phthalate-containing plastics were avoided. Approximately 1 g of each cosmetic sample was weighed into a 40-ml beaker, mixed thoroughly with about 3 g of Celite, and then transferred to a 15-ml extraction tube containing a filter disk. The sample/Celite mixture was covered with a second filter disk and compacted firmly with a stirring rod. The prepared column was eluted with sufficient hexane to obtain 10 ml of extract in a volumetric flask. The extraction flask was mixed well prior to HPLC analysis.

HPLC ANALYSES

HPLC analyses were carried out on an Agilent 1100 series HPLC, equipped with a quaternary pumping system, an in-line vacuum degasser, a variable wavelength diode array UV-visible absorbance detector, a 20-µl injection loop, and a personal computer with HP Chemstation software.

Chromatographic separation was achieved using a Whatman Partisil ODS-3 5-µm guard column (7.5 mm by 4.6 mm ID) and a Whatman Partisil ODS-3 5-µm analytical

column (250 mm by 4.6 mm ID), both obtained from Altech Chromatography (Deerfield, Illinois). The analytical column was housed in a thermostatted column compartment at room temperature. Chromatographic separation was achieved using a solvent program starting initially at 50% water, 34% acetonitrile, 13% 2-propanol, and 3% methanol that was changed linearly over 35 minutes to 15% water, 55% acetonitrile, 25% 2-propanol, and 5% methanol, and held at the final composition for an additional ten minutes. The mobile phase flow rate was 1.0 ml/min. The mobile phase was gradually returned to the initial mobile phase composition over a period of ten minutes. Phthalate esters were detected at 230 nm.

QUANTITATION

Four-point calibration curves were prepared for each phthalate ester. Phthalates were identified in sample extracts by comparing HPLC retention times with standards, and quantitated using the standard calibration curve for each phthalate ester. Sample extracts were diluted as necessary to confirm that the concentrations were in the linear range of the calibration curve. Calculated phthalate ester concentrations of 10 ppm or less were recorded as not detected.

RECOVERY

The recovery of phthalate esters from cosmetic products was determined by fortifying products with 100 and 1,000 ppm of each ester followed by extraction and HPLC analysis as described above.

RESULTS AND DISCUSSION

In this study, a rapid method for the determination of five phthalate esters was developed and validated. For most cosmetic sample extracts, each phthalate ester was completely separated from other components by HPLC and could be quantitated unambiguously. To further confirm that observed peaks were due to phthalate esters and not impurities, UV spectra of chromatographic peaks were evaluated to determine peak purity. For DMP it was generally necessary to use peak UV spectra to distinguish DMP from other compounds eluting near the retention time of DMP.

HPLC calibration curves obtained for BBP, DBP, and DEHP were found to be linear over the concentration range of 0.001 mg/ml to 1 mg/ml. The calibration curve for DMP was linear from 0.001 mg/ml to 0.3 mg/ml, while DEP was linear from 0.001 mg/ml to 0.6 mg/ml. All regression correlation coefficients were better than 0.995. The limit of quantitation (LOQ) ranged from 1 to 10 ppm at ten times baseline noise. Figure 1 shows the chromatographic separation of the five phthalate esters and typical chromatograms of cosmetic extracts.

The presence of phthalate esters in solvents, laboratory equipment, and plastic materials has been reported by other investigators. To assure accurate quantitation of phthalate esters in the cosmetic products examined, laboratory equipment and glassware were carefully washed and thoroughly rinsed with water and ethanol before use. The HPLC

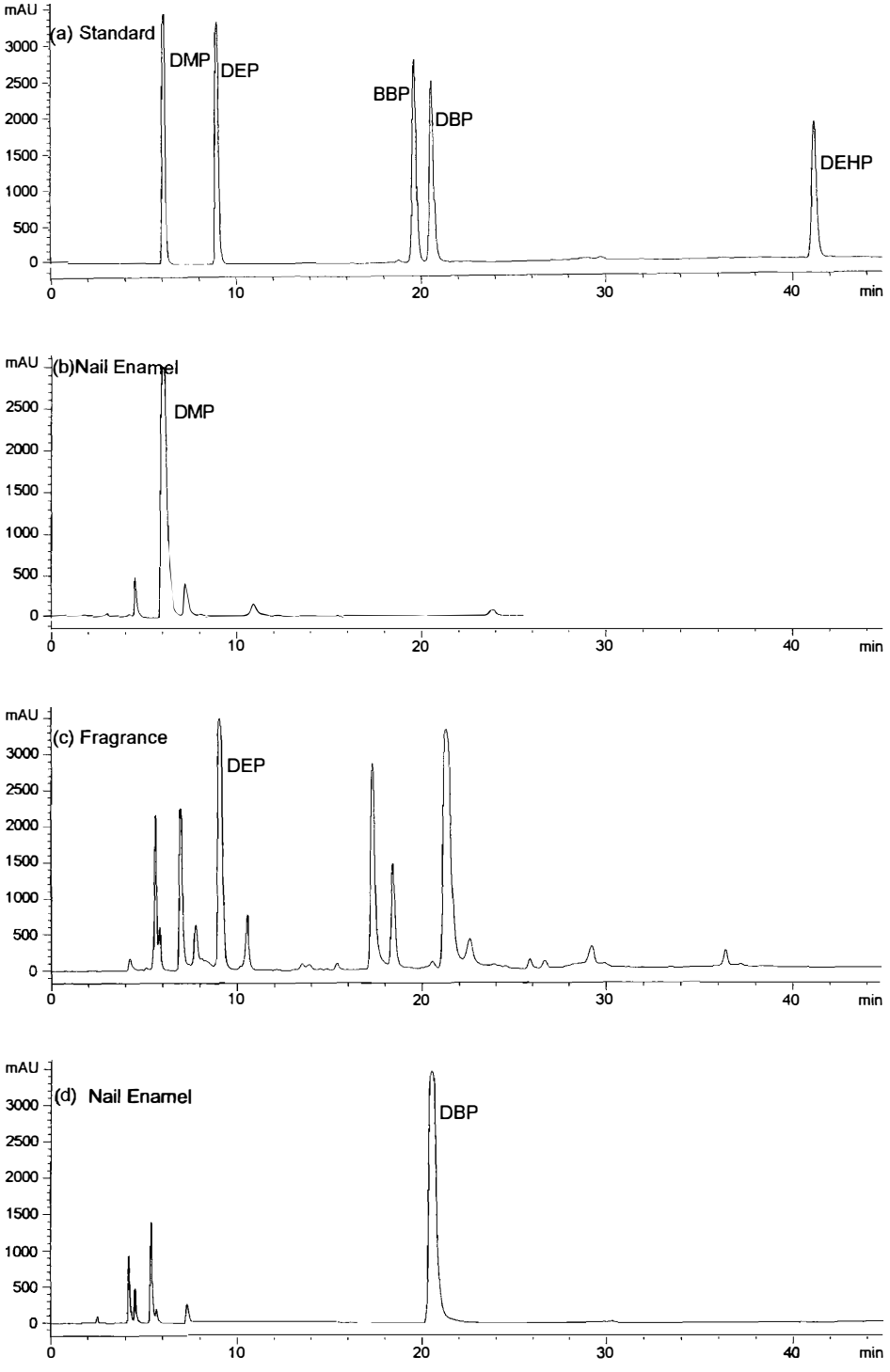

Figure 1. HPLC chromatograms of (a) phthalate ester standards, (b) nail enamel, (c) fragrance product, and (d) nail enamel.

system was flushed with mobile phase at the beginning of each day, and after each injection the syringe was thoroughly washed with ethanol. Since DEHP is particularly persistent in the environment, and is widely used as a plasticizer, plastic materials were not used to process samples. Solvent blanks were run to confirm the absence of phthalates. No chromatographic responses were observed at the retention times of any of the phthalate esters.

Since a wide range of phthalate ester levels are present in cosmetic products, the quantity of sample extracted was occasionally varied, depending on the expected level of phthalate ester in the product. For cosmetic products having an unknown concentration of phthalate ester, a preliminary analysis was made to determine the approximate level and to confirm the absence of significant chromatographic interference. Then an appropriate sample size was selected for analysis. For most products 1 g of sample was analyzed.

Method accuracy was evaluated by performing recovery experiments from two hairspray products, a hand lotion, and an antiperspirant. Each product was fortified with the five phthalate esters at levels of 100 and 1000 ppm. The results are shown in Table I. Recoveries of the five phthalate esters ranged from 73% to 112%. Average recoveries for DMP, DEP, BBP, DBP, and DEHP in the four products were 91%, 95%, 101%, 101%, and 92%, respectively.

A survey of a variety of consumer cosmetic products for phthalate esters was conducted. Products were purchased from local stores in the Washington, DC, area, including hair care products, deodorants, lotions and creams, nail products, fragrances, and body washes. An attempt was made to purchase many of the same products analyzed by the EWG (15) to confirm the reported phthalate ester levels. The results of the analysis of 48 cosmetic products is shown in Table II. Levels less than 10 ppm are reported as not detected. Sixty-seven percent of the products analyzed contained at least one phthalate ester, while hair sprays, deodorants, nail products, and hair mousse contained two or more phthalates. The highest phthalate ester concentrations were found in nail products, with levels observed up to 59,815 ppm. DEP was the most common phthalate ester found; it was present in twenty-seven products. DBP was found in ten products, while DEHP was not found in any product tested. With few exceptions, there was very good agreement between the phthalate ester levels found and those reported by the EWG. Differences observed may be due to lot variations.

Table I
Recovery of Phthalate Esters from Cosmetic Products*

Product	Fortification level (ppm)	Percent recovery				
		DMP	DEP	BBP	DBP	DEHP
Hairspray A	100	90	99	99	95	84
	1000	99	103	109	112	108
Hand lotion	100	89	88	97	102	73
	1000	92	94	95	95	102
Antiperspirant	100	83	94	100	103	81
	1000	88	95	100	99	101
Hairspray B	100	89	90	105	105	84
	1000	94	98	103	100	103

* Single determination at each level.

Table II
Phthalate Esters in Thirteen Commercial Cosmetic Products (PPM)

Product	No. of products	DMP[1]	DEP[2]	BBP[3]	DBP[4]	DEHP[5]
Body lotion	1	ND[6]	142	ND	ND	ND
Hairspray	8	ND	81, 118, 178, 204	43	16, 38, 54	ND
Deodorant	9	ND	38, 56, 57, 111, 681, 805, 2933	ND	104	ND
Fragrance	5	ND	5486, 8851, 9081, 14124, 38663	ND	ND	ND
Skin lotion	3	ND	84	ND	ND	ND
Hair gel	5	ND	53, 67	ND	ND	ND
Hair mousse	5	ND	31, 56, 75, 128	ND	31, 43	ND
Body wash	3	ND	200, 325	ND	ND	ND
Shampoo	1	ND	ND	ND	ND	ND
Hand cream	2	ND	27	ND	ND	ND
Nail enamel	6	58, 143, 15395	1136	107	25, 742, 46463, 59815	ND

[1] Dimethyl phthalate.
[2] Diethyl phthalate.
[3] Benzylbutyl phthalate.
[4] Dibutyl phthalate.
[5] Diethylhexyl phthalate.
[6] None detected (<10 ppm).
[7] Number of products containing the phthalate.

The source of phthalate esters in most cosmetic products is most likely the fragrance ingredient. Phthalate esters were only included on the ingredient statements of some of the nail products included in this survey. Individual fragrance ingredients are not required to be included in cosmetic product labeling (64).

The Cosmetic Ingredient Review (CIR) Expert Panel, an independent panel of scientists that has been reviewing the safety of cosmetic raw materials since 1976, has reviewed the safety of several phthalate esters used in cosmetic products. In the first review, conducted in 1985, the CIR concluded that DMP, DBP, and DEP were safe in cosmetic products at levels up to 5%, 25%, and 50%, respectively (65). In a separate review of the safety of BBP, the CIR concluded that BBP is safe at concentrations less than 1% (66). In 2003, the CIR rereviewed the safety of phthalate esters in cosmetic products in light of reports of phthalate metabolites in human urine, and affirmed their original conclusions that the levels used in cosmetic products were safe. From 1998 to 2000, an expert panel convened by the NTP concluded that reproductive risks from exposure to phthalate esters were minimal to negligible in most cases (67). The NTP has concluded that food is the primary source of human exposure to DBP (33). In the European Union, the Scientific Committee on Cosmetics and Non-Food Products reviewed the safety of DEP and concluded it was safe in cosmetic products (68); however, the committee concluded that DBP should not be intentionally added to cosmetics (69).

The significance of phthalate ester exposure from cosmetics compared to exposure from food, water, air, and plastic materials is difficult to assess. Exposure from pharmaceuticals must also be factored in, since high urinary levels of the metabolite of DBP have

been traced to the use of drugs (70). Our survey of cosmetic products found that the highest levels of phthalate esters were present in nail and fragrance products. Products such as nail polish harden rapidly after application, and so phthalate ester absorption through the nail is likely to be significantly inhibited. Exposure to phthalate esters from products such as soaps, shampoos, and conditioners that are washed off the skin soon after application will also be very low, due to limited contact time with the skin. For cosmetic products that are left on the skin, exposure is a function of the area of skin exposed to the product and the absorption rate, and it has been shown that phthalate ester absorption rates through human skin are slow compared to those of rodents (71).

Since our 2002 survey was conducted, the FDA has observed that some cosmetic products are being reformulated to remove phthalate esters. The FDA will continue to monitor and evaluate all available data to assure that phthalate ester levels in cosmetic products are not a health concern. The Federal Food, Drug and Cosmetic Act does not provide for premarket approval of cosmetic products, and the standard for regulatory action requires that the agency prove a product is adulterated or harmful under conditions of use. Based on the safety and toxicity data currently available, the agency has concluded that there is no basis upon which to take regulatory action at this time. If the FDA determines that a health hazard exists, the agency will advise the public and will consider its regulatory options.

REFERENCES

(1) H. Fromme, T. Küchler, T. Otto, K. Pilz, J. Müller, and A. Wenzel, Occurrence of phthalates and bisphenol A and F in the environment, *Water Res.,* **36,** 1429–1438 (2002).
(2) J.-D. Berset and R. Etter-Holzer, Determination of phthalates in crude extracts of sewage sludges by high-resolution capillary gas chromatography with mass spectrometric detection, *J. AOAC Internat.,* **84,** 383–391 (2001).
(3) A. Peñalver, E. Pocurull, F. Borrull, and R. M. Marcé, Comparison of different fibers for the solid-phase microextraction of phthalate esters from water, *J. Chromatogr. A,* **922,** 377–384 (2001).
(4) A. Peñalver, E. Pocurull, F. Borrull, and R. M. Marcé, Determination of phthalate esters in water samples by solid-phase microextraction and gas chromatography with mass spectrometric detection, *J. Chromatogr. A,* **872,** 191–201 (2000).
(5) J. J. Adibi, F. P. Perera, W. Jedrychowski, D. E. Camann, D. Barr, R. Jacek, and R. M. Whyatt, Prenatal exposures to phthalates among women in New York City and Krakow, Poland, *Environ. Health Perspect.,* **111,** 1719–1722 (2003).
(6) H. Toda, K. Sako, Y. Yagome, and T. Nakamura, Simultaneous determination of phosphate esters and phthalate esters in clean room air and indoor air by gas chromatography–mass spectrometry, *Anal. Chim. Acta,* **519,** 213–218 (2004).
(7) O. W. Lau and S.-K. Wong, Determination of plasticisers in food by gas chromatography–mass spectrometry with ion-trap mass detection, *J. Chromatogr. A,* **737,** 338–342 (1996).
(8) J. H. Petersen, Survey of di-(2-ethylhexyl)phthalate plasticiser contamination of retail Danish milks, *Food Add. Contam.,* **8,** 701–706 (1991).
(9) I. Tomita, Y. Naramura, and Y. Yagi, Phthalic acid esters in various foodstuffs and biological materials, *Ecotoxicol. Environ. Safety,* **1,** 275–287 (1977).
(10) G. Di Bella, M. Saitta, M. Pellegrino, F. Salvo, and G. Dugo, Contamination of Italian citrus essential oils: Presence of phthalate esters, *J. Agric. Food Chem.,* **47,** 1009–1012 (1999).
(11) T. Dine, M. Luychx, M. Cazin, J. C. Cazin, and F. Goudaliez, Rapid determination by high performance liquid chromatography of di-2-ethylhexyl phthalate in plasma stored in plastic bags, *Biomed. Chromatogr.,* **5,** 94–97 (1991).
(12) J. Vessman and G. Rietz, Determination of di(ethylhexyl)phthalate in human plasma and plasma proteins by electron capture gas chromatography, *J. Chromatogr.,* **100,** 153–163 (1974).

(13) H. G. Wahl, A. Hoffmann, H.-U. Häring, and H. M. Liebich, Identification of plasticizers in medical products by a combined direct thermodesorption-cooled injection system and gas chromatography–mass spectrometry, *J. Chromatogr.,* **847,** 1–7 (1999).
(14) M. Fujii, N. Shinohara, A. Lim, T. Otake, K. Kumagai, and Y. Yanagisawa, A study on emission of phthalate esters from plastic materials using a passive flux sampler, *Atmospheric Environ.,* **37,** 5495–5504 (2003).
(15) J. Houlihan, C. Brody, and B. Schwan, "Not Too Pretty: Phthalates, Beauty Products, & the FDA," a report published by the Environmental Working Group, Coming Clean, and Health Care Without Harm, July 8, 2002 (*http://www.ewg.org/reports_content/nottoopretty/NotTooPretty_final.pdf*).
(16) B. C. Blount, M. J. Silva, S. P. Caudil, L. L. Needham, J. L. Pirkle, E. J. Sampson, G. W. Lucier, R. J. Jackson, and J. W. Brock, Levels of seven urinary phthalate metabolites in a human reference population, *Environ. Health Perspect.,* **108,** 979–982 (2000).
(17) J. A. Hoppin, J. W. Brock, B. J. Davis, and D. D. Baird, Reproducibility of urinary phthalate metabolites in first morning urine samples, *Environ. Health Perspect.,* **110,** 515–518 (2002).
(18) J. W. Koo, F. Parham, M. C., Kohn, S. A. Masten, J. W. Brock, L. L. Needham, and C. J. Portier, The association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population, *Environ. Health Perspect.,* **110,** 405–410 (2002).
(19) H. Koch, M. Holger, B. Rossbach, H. Drexler, and J. Angerer, Internal exposure of the general population to DEHP and other phthalates—Determination of secondary and primary phthalate monoester metabolites in urine, *Environ. Res.,* **93,** 177–185 (2003).
(20) A. M. Calafat, A. Slakman, S. Ryan, J. Manori, A. R. Herbert, and L. L. Needham, Automated solid phase extraction and quantitative analysis of human milk for 13 phthalate metabolites, *J. Chromatogr. B,* **805,** 49–56 (2004).
(21) M. C. Kohn, F. Parham, S. A. Masten, C. J. Portier, M. D. Shelby, J. W. Brock, and L. L. Needham, Human exposure to phthalate esters, *Environ. Health Perspect.,* **108,** A440–A442 (2000).
(22) M. Ema, E. Miyawaki, and K. Kawashima, Further evaluation of developmental toxicity of di-n-butyl phthalate following administration during late pregnancy in rats, *Toxicol. Lett.,* **98,** 87–93 (1998).
(23) F. A. Arcadi, C. Costa, C. Imperatore, A. Marchese, A. Rapisarda, M. Salemi, G. R. Trimarchi, and G. Costa, Oral toxicity of bis(2-ethylhexyl) phthalate during pregnancy and suckling in the Long-Evans rat, *Food Chem. Toxicol.,* **36,** 963–970 (1998).
(24) M. Ema, E. Miyawaki, and K. Kawashima, Effects of dibutyl phthalate on reproductive function in pregnant and pseudopregnant rats, *Reprod. Toxicol.,* **14,** 13–19 (2000).
(25) M. Ema, E. Miyawaki, and K. Kawashima, Critical period for adverse effects on development of reproductive system in male offspring of rats given di-n-butyl phthalate during late pregnancy, *Toxicol. Lett.,* **111,** 271–278 (2000).
(26) M. Ema and E. Miyawaki, Effects of monobutyl phthalate on reproductive function in pregnant and pseudopregnant rats, *Reprod. Toxicol.,* **15,** 261–267 (2001).
(27) M. Ema and E. Miyawaki, Adverse effects on development of the reproductive system in male offspring of rats given monobutyl phthalate, a metabolite of dibutyl phthalate, during late pregnancy, *Reprod. Toxicol.,* **15,** 189–194 (2001).
(28) E. Mylchreest, R. C. Cattley, and P. M. D. Foster, Male reproductive tract malformations in rats following gestational and lactational exposure to di(n-butyl) phthalate: An antiandrogenic mechanism?, *Toxicol. Sci.,* **43,** 47–60 (1998).
(29) M. Ema and E. Miyawaki, Effects on development of the reproductive system in male offspring of rats given butyl benzyl phthalate during late pregnancy, *Reprod. Toxicol.,* **16,** 71–76 (2002).
(30) M. Ema, E. Miyawaki, A. Hirose, and E. Kamata, Decreased anogenital distance and increased incidence of undescended testes in fetuses of rats given monobenzyl phthalate, a major metabolite of butyl benzyl phthalate, *Reprod. Toxicol.,* **17,** 407–412 (2003).
(31) S. D. Gangolli, Testicular effects of phthalate esters, *Environ. Health Perspect.,* **45,** 77–84 (1982).
(32) T. Ichimura, M. Kawamura, and A. Mitani, Co-localized expression of FasL, Fas, Caspase-3 and apoptotic DNA fragmentation in mouse testis after oral exposure to di(2-ethylhexyl)phthalate, *Toxicology,* **194,** 35–42 (2003).
(33) R. Kavlock, K. Boekelheide, R. Chapin, M. Cunningham, E. Faustman, P. Foster, M. Golub, R. Henderson, I. Hinberg, and R. Little, NTP Center for the Evaluation of Risks to Human Reproduction: Phthalates expert panel report on the reproductive and developmental toxicity of di-n-butyl phthalate, *Reprod. Toxicol.,* **16,** 489–527 (2002).
(34) R. Kavlock, K. Boekelheide, R. Chapin, M. Cunningham, E. Faustman, P. Foster, M. Golub, R.

Henderson, I. Hinberg, and R. Little, NTP Center for the Evaluation of Risks to Human Reproduction: Phthalates expert panel report on the reproductive and developmental toxicity of butyl benzyl phthalate, *Reprod. Toxicol.,* **16,** 453–487 (2002).
(35) K.-Y. Lee, M. Shibutani, H. Takagi, N. Kato, S. Takigami, C. Uneyama, and M. Hirose, Diverse developmental toxicity of di-n-butyl phthalate in both sexes of rat offspring after maternal exposure during the period from late gestation through lactation, *Toxicology,* **203,** 221–238 (2004).
(36) R. N. Wine, L.-H. Li, L. H. Barnes, D. K. Gulati, and R. E. Chapin, Reproductive toxicity of di-n-butylphthalate in a continuous breeding protocol in Sprague-Dawley rats, *Environ. Health Perspect.,* **105,** 102–107 (1997).
(37) J. Doull, R. Cattley, C. Elcombe, B. G. Lake, J. Swenberg, C. Wilkenson, G. Williams, and M. A. van Gemert, A cancer risk assessment of di(2-ethylhexyl)phthalate: Application of the new U.S. EPA risk assessment guidelines, *Regul. Toxicol. Pharmacol.,* **29,** 327–357 (1999).
(38) R. L. Melnick, Is peroxisome proliferation an obligatory precursor step in the carcinogenicity of di(2-ethylhexyl)phthalate (DEHP)?, *Environ. Health Perspect.,* **109,** 437–442 (2001).
(39) S. M. Duty, N. P. Singh, M. J. Silva, D. B. Barr, J. W. Brock, L. Ryan, R. F. Herrick, D. C. Christiani, and R. Hauser, The relationship between environmental exposures to phthalates and DNA damage in human sperm using the neutral comet assay, *Environ. Health Perspect.,* **111,** 1164–1169 (2003).
(40) Y. Tomonari, Y. Kurata, T. Kawasuso, R. David, G. Gans, M. Tsuchitani, and M. Katoh, Testicular toxicity study of di(2-ethylhexyl) phthalate (DEHP) in juvenile common marmoset, *Toxicologist,* **72,** 385 (2003).
(41) FDA, 2004 priorities (*http://www.cfsan.fda.gov/~dms/cfsan404.html*).
(42) R. Battle and C. Nerin, Application of single-drop microextraction to the determination of dialkyl phthalate esters in food simulants, *J. Chromatogr. A,* **1045,** 29–35 (2004).
(43) G. Prokůpková, K. Holadová, J. Poustka, and J. Hajšlová, Development of a solid-phase microextraction method for the determination of phthalic acid esters in water, *Anal. Chim. Acta,* **457,** 211–233 (2002).
(44) O. A. Teirlynck and M. T. Rosseel, Determination of di- and mono (2-ethylhexyl) phthalate in plasma by gas chromatography, *J. Chromatogr.,* **342,** 399–405 (1985).
(45) K. S. Andersen and J. Lam, Simple and direct method for quantitative gas chromatographic determination of di(2-ethylhexyl) phthalate in edible oils, *J. Chromatogr.,* **169,** 101–106 (1979).
(46) Y.-Q. Cai, G.-B. Jiang, J.-F. Liu, and Q.-X. Zhou, Multi-walled carbon nanotubes packed cartridge for the solid-phase extraction of several phthalate esters from water samples and their determination by high performance liquid chromatography, *Anal. Chim. Acta,* **494,** 149–156 (2003).
(47) K. Mitani, S. Narimatsu, F. Izushi, and H. Kataoka, Simple and rapid analysis of endocrine disruptors in liquid medicines and intravenous injection solutions by automated in-tube solid-phase microextraction/high performance liquid chromatography, *J. Pharm. Biomed. Anal.,* **32,** 469–478 (2003).
(48) K. Kambia, T. Dine, B. Gressier, A.-F. Germe, M. Luyckx, C. Brunet, L. Michaud, and F. Gottrand, High-performance liquid chromatographic method for the determination of di(2-ethylhexyl) phthalate in total parenteral nutrition and in plasma, *J. Chromatogr. B,* **755,** 297–303 (2001).
(49) S. Jara, C. Lysebo, T. Greibrokk, and E. Lundanes, Determination of phthalates in water samples using polystyrene solid-phase extraction and liquid chromatography quantification, *Anal. Chim. Acta,* **407,** 165–171 (2000).
(50) L. Brossa, R. M. Marcé, F. Borrull, and E. Pocurull, Application of on-line solid-phase extraction–gas chromatography–mass spectrometry to the determination of endocrine disruptors in water samples, *J. Chromatogr. A,* **963,** 287–294 (2002).
(51) R. Chaler, L. Canton, M. Vaquero, and J. O. Grimalt, Identification and quantification of n-octyl esters of alkanoic and hexanedioic acids and phthalates as urban wastewater markers in biota and sediments from estuarine areas, *J. Chromatogr. A,* **1046,** 203–210 (2004).
(52) E. Cortazar, O. Zuloaga, J. Sanz, J. C. Raposo, N. Etxebarria, and L. A. Fernandez, MultiSimplex optimisation of the solid-phase microextraction–gas chromatographic–mass spectrometric determination of polycyclic aromatic hydrocarbons, polychlorinated biphenyls and phthalates from water samples, *J. Chromatogr. A,* **978,** 165–175 (2002).
(53) K. Luks-Betlej, P. Popp, B. Janoszka, and H. Paschke, Solid-phase microextraction of phthalates from water, *J. Chromatogr. A,* **938,** 93–101 (2001).
(54) T. Niino, T. Ishibashi, T. Itho, S. Sakai, H. Ishiwata, T. Yamada, and S. Onodera, Simultaneous determination of phthalate di- and monoesters in poly(vinylchloride) products and human saliva by gas chromatography–mass spectrometry, *J. Chromatogr. B,* **780,** 35–44 (2002).

(55) C. Y. Chen, A. V. Ghule, W. Y. Chen, C. C. Wang, Y. S. Chiang, and Y. C. Ling, Rapid identification of phthalates in blood bags and food packaging using ToF-SIMS, *Appl. Surface Sci.,* **231–232,** 447–451 (2004).
(56) A. O. Earls, I. P. Axford, and J. H. Braybrook, Gas chromatography–mass spectrometry determination of the migration of phthalate plasticisers from polyvinyl chloride toys and childcare articles, *J. Chromatogr. A,* **983,** 237–246 (2003).
(57) B. C. Blount, K. E. Milgram, M. J. Silva, N. A. Malek, J. A. Reidy, L. L. Needham, and J. W. Brock, Quantitative detection of eight phthalate metabolites in human urine using HPLC-APCI-MS/MS, *Anal. Chem.,* **72,** 4127–4134 (2000).
(58) H. M. Koch, L. M. Gonzalez-Reche, and J. Angerer, On-line clean-up by multidimensional liquid chromatography–electrospray ionization tandem mass spectrometry for high throughput quantification of primary and secondary phthalate metabolites in human urine, *J. Chromatogr. B,* **784,** 169–182 (2003).
(59) M. J. Silva, A. R. Slakman, J. A. Reidy, J. L. Preau, Jr., A. R. Herbert, E. Samandar, L. L. Needham, and A. M. Calafat, Analysis of human urine for fifteen phthalate metabolites using automated solid-phase extraction, *J. Chromatogr. B,* **805,** 161–167 (2004).
(60) K. Inoue, T. Higuchi, F. Okada, H. Iguchi, Y. Yoshimura, A. Sato, and H. Nakazawa, The validation of column-switching LC/MS as a high-throughput approach for direct analysis of di(2-ethylhexyl) phthalate released from PVC medical devices in intravenous solution, *J. Pharm. Biomed. Anal.,* **31,** 1145–1152 (2003).
(61) W. Hancock, B. A. Rose, and D. D. Singer, The determination of diethyl phthalate in cosmetic preparations, *Analyst,* **91,** 449–454 (1966).
(62) E. W. Godly and A. E. Mortlock, The determination of di-n-alkyl phthalates in cosmetic preparations by gas-liquid chromatography, *Analyst,* **98,** 493–501 (1973).
(63) R. L. Yates and D. C. Havery, Determination of phenol, resorcinol, salicylic acid, and α-hydroxy acids in cosmetic products and salon preparations, *J. Cosmet. Sci.,* **50,** 315–325 (1999).
(64) *Code of Federal Regulations,* 21, 701.3(a).
(65) Cosmetic Ingredient Review: Final report on the safety assessment of dibutyl phthalate, dimethyl phthalate, and diethyl phthalate, *J. Am. Coll. Toxicol.,* **4,** 267–303 (1985).
(66) Cosmetic Ingredient Review: Final report on the safety assessment of butyl benzyl phthalate, *J. Am. Coll. Toxicol.,* **11,** 1–23 (1992).
(67) R. H. McKee, J. H. Butala, R. M. David, and G. Gans, NTP center for the evaluation of risks to human reproduction reports on phthalates: Addressing the data gaps, *Reprod. Toxicol.,* **18,** 1–22 (2004).
(68) Scientific Committee on Cosmetics and Non-food Products (*http://europa.eu.int/comm/health/ph_risk/committees/sccp/documents/out246_en.pdf*).
(69) Scientific Committee on Cosmetics and Non-food Products (*http://europa.eu.int/comm/health/ph_risk/committees/sccp/documents/out287_en.pdf*).
(70) R. Hauser, S. Duty, L. Godfrey-Bailey, and A. M. Calafat, Medications as a source of human exposure to phthalates, *Environ. Health Perspect.,* **112,** 751–753 (2004).
(71) R. C. Scott, P. H. Dugard, J. D. Ramsay, and C. Rhodes, In vitro absorption of some o-phthalate diesters through human and rat skin, *Environ. Health Perspect.,* **74,** 223–227 (1987).