

Determination of Phthalates by Binary HPLC

Introduction:

Phthalates are in the majority of cases used as plasticizers in flexible polyvinylchloride products. The chemical industry produces according to the “European Council for Plasticizers and Intermediates”(ECPI) about 1 millions every year only for Western Europe. Although their vapor pressures are generally low, phthalates may occur including a vapor phase. Their general lipophilic character influences the leaching and environmental partitioning characteristics. Phthalates are evaporated from consumer products or find their way into the environment by abrasion from PVC particles. It becomes obvious that humans get into contact with these substances easily. Potential pathways of exposure are ingestion, inhalation, intravenous injection and skin absorption. Consumer products containing phthalates can result in human exposures through direct contact and use, by leaching into other products, or via general environmental contamination. Phthalates and their metabolites can be found in every human today, for example in urine or blood. Not at least caused by their adverse health effects, phthalates have to be monitored critically. In this work, eight of the most commonly used phthalates, namely Benzyl benzoate, butyl benzyl phthalate (BBP), dibutyl phthalate (DBP), dihexyl phthalate(DHP), di-(2-ethylhexyl) Phthalate (DEHP), di-n-octyl phthalate (DNOP), di-isononyl phthalate (DINP) and diisodecyl phthalate (DIDP) are separated. Their chemical structures are shown in figure 1. The EU classified DEHP, DBP and BBP as toxic to reproduction and banned them especially from baby products. In many cases they can be replaced by DIDP and DINP for example, which are until now not regarded as toxic. Baby products are an exception, where these softeners are

also forbidden for preventative reasons. DINP and DIDP are under suspicion for quickly spreading in the environment and accumulating in organisms. For this reason, their entry in the environment has to be inhibited. The German “Umwelt Bundesamt” suggests replacing all phthalate containing materials like flexible PVC little by little with phthalate free materials like polyethylene and polypropylene where it is possible.

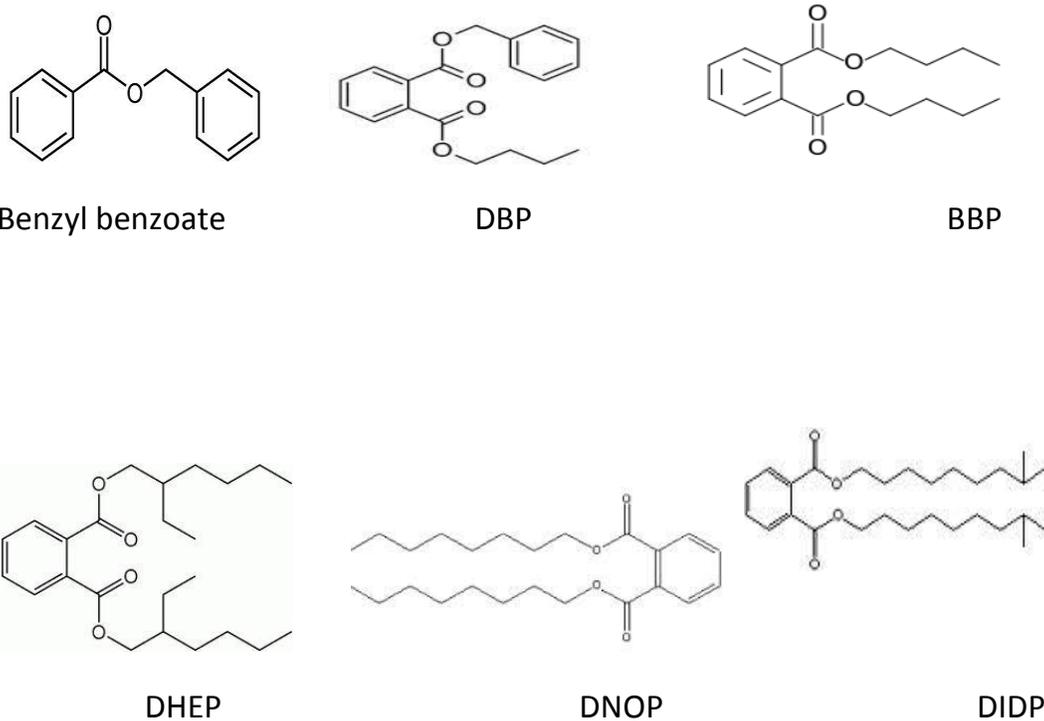


Figure 1: Structure of analyzed Phalates

Experimental Sample Preparation:

According to the United States Consumer Product Safety Commission (CPSC) Test Method

CPSC-CH-C1001-09.1, phthalates can be extracted from consumer products after comminution. An amount of 0.05 g of the crushed sample is collected in a glass vial and 5 ml of THF are added. The vial is shaken until the sample is dissolved what may take 30 min up to 2 h depending on the material. Polymers are precipitated using 10 ml of hexane or methanol in combination with cooling. When the polymers have settled, the solution is filtered through a 0.45 μm PTFE filter, evaporated and then diluted again with acetonitrile. After optionally adding an internal standard and dilution depending on the phthalate concentration the sample can be analyzed by HPLC.

Experimental Preparation of Standard Solution:

In this work a stock solution was made by weighing out the single compounds, dissolving and mixing them in concentrations noted in table 1. Benzylbenzoate can act as an internal standard when samples are prepared. After dilution 1:10 with water/acetonitrile 15:85(v/v) the standard solution is ready for analysis by HPLC.

COMPONENT	Concentration (mg/ml)
Benzylbenzoate	40.0
BBP	200.1
DBP	200.6
DHP	194.2
DEHP	206.0
DNOP	197.1
DINP	231.1
DIDP	211.5

Table 1: Conc. Of Standard Solution

Method Parameters:

Column: Biochrome 100-3, C18 H, 250x3mm

Eluent A: Water: Acetonitrile (15:85)v/v

Eluent B: Acetonitrile

Flow rate: 0.6 ml/min

Injection Volume: 2 μ L

Standard Column Temperature: 30°C System

Pressure: approx 235 bar

Detection: UV at 225nm

Analysis time: 21 min Run Time: 45 min

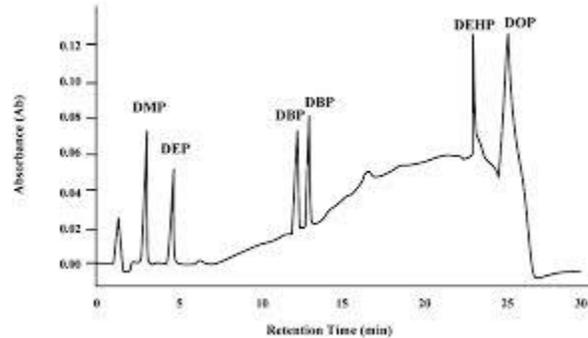


Figure 2: Chromatogram of Standard Solution 2MI

Determination of Pentachlorophenol & Tetrachlorophenol by ATL Binary HPLC 3000

Apparatus:

A high performance liquid chromatograph equipped with sample injector, a cyano bonded phase HPLC column, a variable wavelength UV detector, and a chart recorder are needed for the analysis. ATL 3000 pump, ATL 3000 UV-Visible detector and a Biochrome 25-cm × 4.6-mm i.d. CN bonded phase column were used in this study. Various sizes of volumetric glassware and pipettes are needed for sample and standard preparations. Three-milliliter (or larger) screw-cap or crimp-type vials are needed. Small brown glass bottles fitted with inert cap liners are needed to store standard solutions. A repetitive dispenser capable of accurately delivering the desorption solution is needed.

Reagents:

HPLC grade methanol and acetonitrile, Reagent grade phosphoric acid, HPLC grade water, a reagent grade standard of pentachlorophenol is required.

Standard preparation:

Prepare a stock solution of pentachlorophenol by accurately weighing approximately 200 mg of the standard and transferring it to a 25-mL volumetric flask. Dilute to volume with methanol. Dilute this stock solution 1/25 to yield an approximate 320 µg/mL solution in methanol. Prepare 1/50, 1/25 and 2/25 dilutions of the 320 µg/mL PCP solution to yield standards which correspond approximately to 0.5, 1, and 2 times the PEL for the recommended sampling conditions.

Sample preparation

Prepare samples for analysis by transferring the entire contents of the sampling tube including glass wool plugs, the resin, and the glass fiber disc into a 4-mL vial. The transfer can best be accomplished if the glass fiber disc is first transferred to the sample vial and the front glass wool plug partially removed. Then with the sampling tube inserted into the vial, use a small spatula or glass stirring rod to force the entire contents of the tube including both glass wool plugs into the vial. Rinse the inside of the sampling tube into the sampling vial with two 1-mL portions of methanol using a 1-mL repetitive dispenser.

Analysis

Prepare a high performance liquid chromatograph for sample analysis using the HPLC conditions listed below.

column:	Biochrome 25-cm × 4.6-mm i.d. CN bonded phase
mobile phase:	40/60 (v/v) acetonitrile/water containing approximately 0.1% by volume of phosphoric acid.
flow rate:	1.3 mL/min
UV detector:	220 nm
injection volume:	25 µL
retention time:	8.1 min

Since column-to-column variations do occur, it is important to ensure that PCP is being separated from the other chlorinated phenols. The injection of a tetra/pentachlorophenol standard mixture will produce baseline separation between pentachlorophenol and the tetrachlorophenol isomers if the

analytical conditions are properly selected. Under these conditions the other chlorinated phenols are not interferences. Interferences: Any compound which has the same retention time as pentachlorophenol is a potential interference. Under the recommended analytical conditions none of the mono-, di-, tri-, or tetrachlorophenols interfered with the analysis. Comparison of peak height ratios of analyte response at two wavelengths for both samples and standards is a valuable confirmatory technique in HPLC. Normal phase HPLC methods and gas chromatographic methods may also be used for sample confirmation. GC/mass spectrometry is an additional confirmatory technique which may be used.

Calculations

Prepare a standard calibration curve of area response versus concentration for PCP by determining the least squares fit equation for the curve. Calculate the analyte concentration in the samples by entering their response values into the equation and solving for the sample concentration. A laboratory data system, or one of many small hand-held calculators, can be used to perform these calculations.

Combine the amount of analyte found on all sections including the sample tube cap. Express results in mg/m³ using the following equation:

$$\text{mg/m}^3 = \frac{(\mu\text{g/mL analyte}) (2\text{-mL desorption volume}) (1 \text{ mg})}{(\text{Air volume in m}^3) (1000 \mu\text{g}) (\text{Solvent adsorption effect})}$$

Note: No solvent adsorption effect correction will need to be applied if standards have been prepared in solutions containing XAD-7 resin.

To convert to ppm at 760 nm and 25°C:

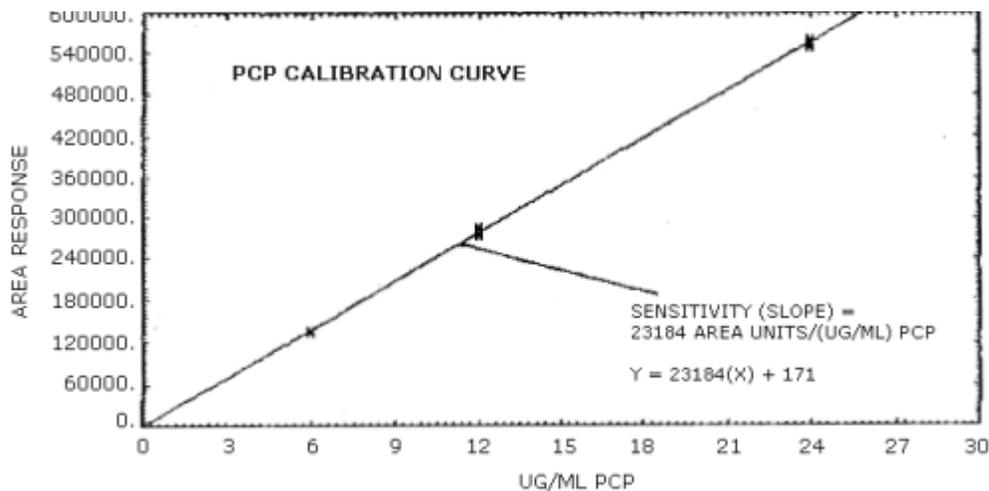
$$\text{ppm} = (\text{mg/m}^3) (24.46) / (\text{MW of analyte})$$

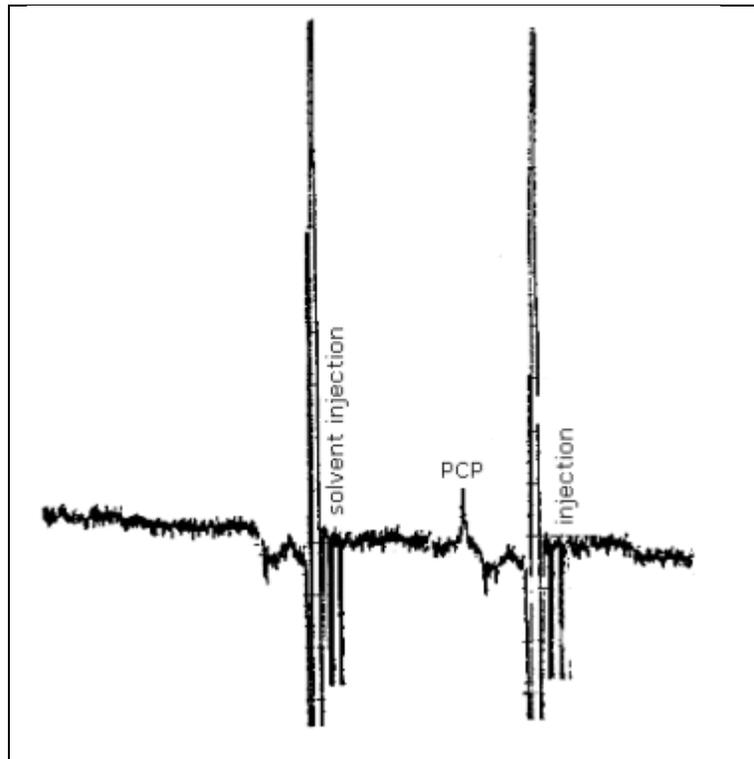
24.46 = the molar volume at 760 nm and 25°C

MW = 266.35 for pentachloropropanol

Safety precautions:

Minimize exposure to pentachlorophenol by performing standard preparations in a well ventilated hood. Avoid all skin contact with pentachlorophenol. Restrict the use of solvents to hoods which provide adequate ventilation. Wear safety glasses in laboratory areas at all times.





Detection limit for PCP