

Application Note

► Rapid determination of phthalates

Category	Environmental analysis
Matrix	Plastics, PVC
Method	UHPLC
Keywords	Phthalates, exposure, consumer products, softener, plasticizer, PVC
Analytes	Benzylbenzoate, butyl benzyl phthalate (BBP), dibutyl phthalate (DBP), dihexyl phthalate (DHP), di-(2-ethylhexyl) phthalate (DEHP), di-n-octyl phthalate (DNOP), di-isononyl phthalate (DINP), diisodecyl phthalate (DIDP)
ID	VEV5, 06/10



Photo: crimex.de –
duck without softeners

Summary

A fast method for the separation and determination of eight commonly used phthalates applying the KNAUER PLATINblue UHPLC System is presented in this application note. Reduction of analysis time from about 35 to less than 5 minutes compared to a conventional HPLC method is achieved by employing the BlueOrchid C18 stationary phase with a 1.8 µm particle size filled in a 2 mm ID column. A binary high pressure gradient instrumentation is applied at a flow rate of 0.5 ml/min in combination with a UV detector.

Introduction

In the majority of cases phthalates are used as plasticizers in flexible polyvinylchloride products. According to the “European Council for Plasticisers and Intermediates” (ECPI) the chemical industry produces about 1 mill. tons every year for West Europe alone. Although their vapor pressure in general is low, phthalates may also occur in the vapor phase. Their generally lipophilic character influences the leaching and environmental partitioning characteristics.¹

Phthalates are evaporated from consumer products or find their way into the environment by abrasion from PVC particles. It is thus apparent that humans can 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. Today phthalates and their metabolites can be found in every human, for example in urine or blood.^{1,2}

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 Umweltbundesamt suggests replacing all phthalate containing materials like flexible PVC step by step with phthalate free materials like polyethylene and polypropylene where it is possible.²

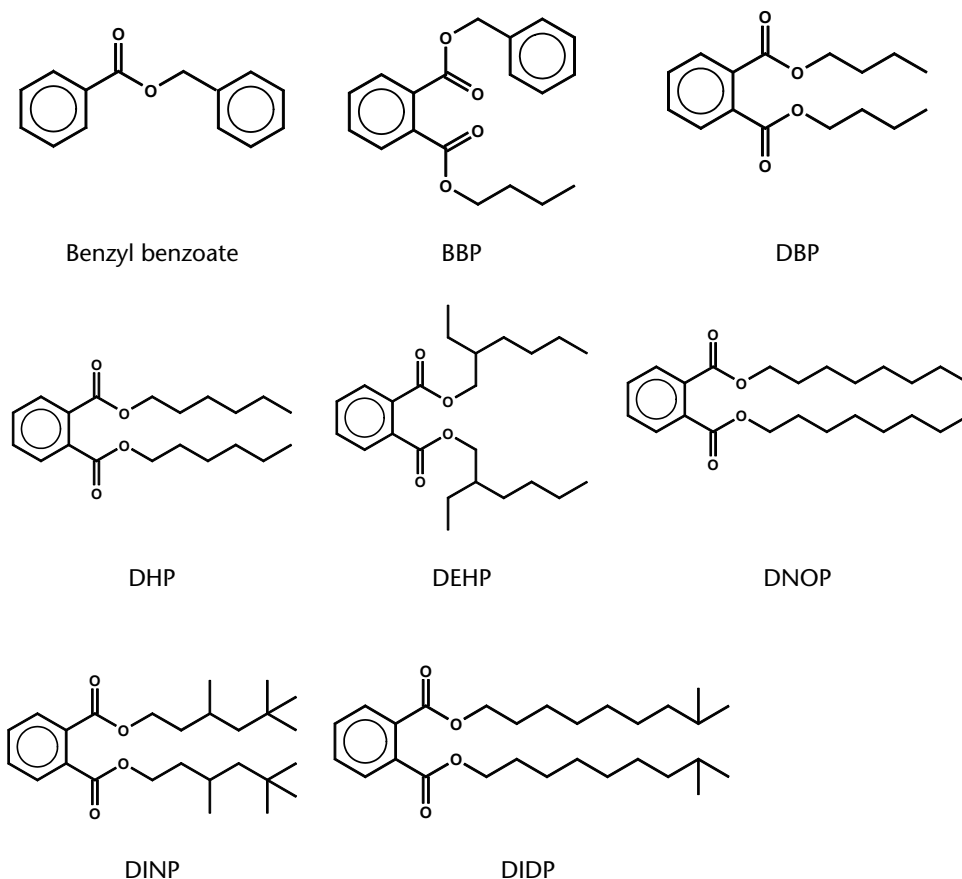


Fig. 1
Structures of analyzed phthalates

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.^{3,4}

Experimental preparation of the 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. Benzyl benzoate 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.

Table 1
Concentration of standard solution

Component	Concentration [mg/ml]
Benzyl benzoate	40.0
BBP	200.1
DBP	200.6
DHP	194.2
DEHP	206.0
DNOP	197.1
DINP	231.1
DIDP	211.5

Method parameters

Column	BlueOrchid C18, 100 x 2 mm ID		
Eluent A	water/acetonitrile 15:85 (v/v)		
Eluent B	acetonitrile		
Gradient	Time [min]	% A	% B
	0.0	100	0
	1.2	100	0
	3.2	0	100
	5.0	0	100
Flow rate	0.5 ml/min		
Injection volume	2 µl standard		
Column temperature	30 °C		
Detection	UV at 225 nm (50 Hz, 0.05 s)		
Analysis time	5 min		
Run time	8 min		

Results

Fig. 2
Chromatogram of the phthalate standard solution (2 µl)

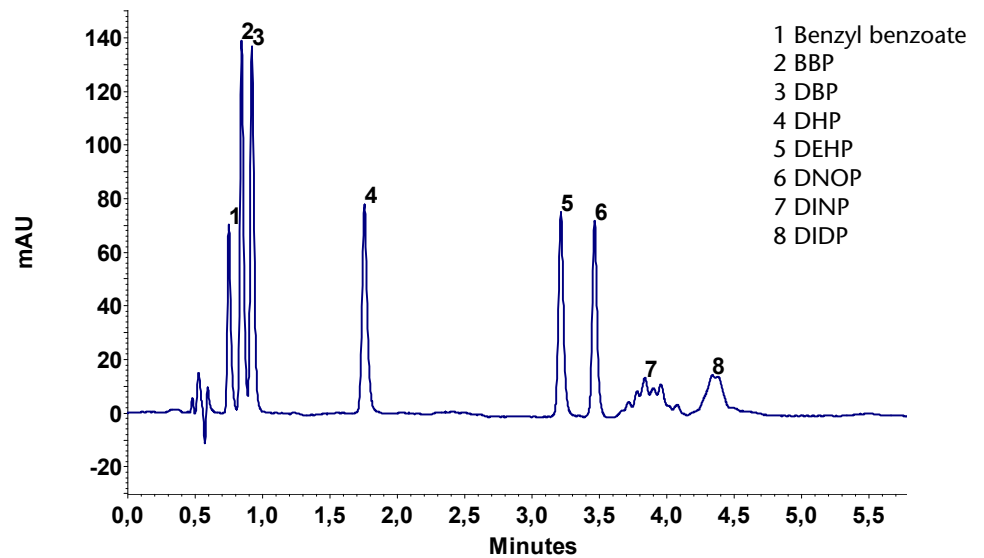
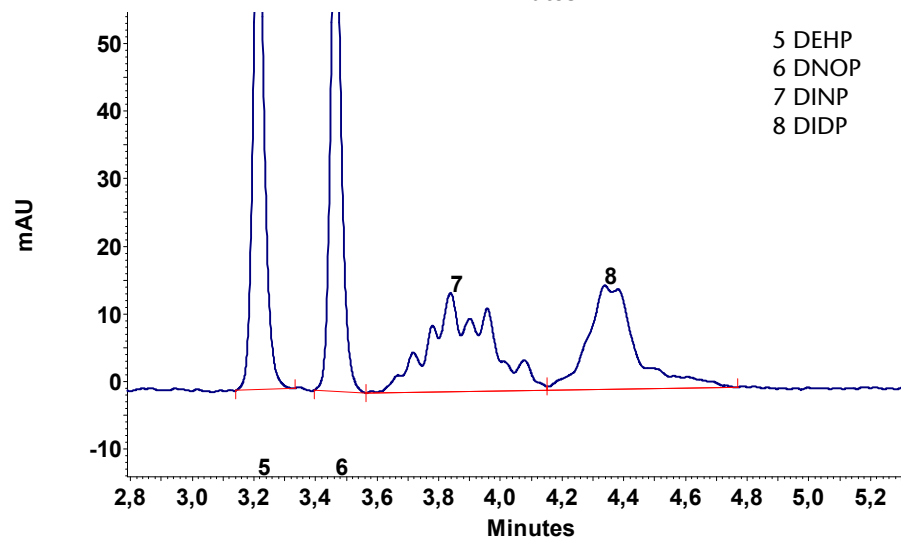


Fig. 3
Chromatogram zoomed in for peaks 7 and 8



Results (continued)

A chromatogram of the phthalate standard is shown in figure 2. All eight substances are baseline separated with resolution values in the range of 1.5 for the critical pair BBP and DBP, up to 21.9 for DEHP. By using a UHPLC method, analysis time could be reduced from nearly 35 to less than 5 min.

When examining the chromatogram shown in figure 2, it becomes obvious that the peak shape for the two substances DINP and DIDP is not as sharp as for the other phthalates and it seems as if these compounds are not eluting in one but in a sum of various peaks. The reason for these phenomena is the quality of the standard. Regarding the chemical structures in figure 1, it is noticeable that DINP and DIDP may occur in different isomeric forms. The quality of a single compound is normally described as "mixture of isomers" by the chemical industry, which cannot easily produce DINP and DIDP free of isomeric forms. In this work, the sum of peaks is regarded as one for the evaluation as shown in figure 3.

Conclusion

This application note describes a very fast method for the determination of eight phthalates that are commonly used by the industry. The easy separation in less than 5 minutes is possible by employing the KNAUER PLATINblue UHPLC system, a BlueOrchid C18 stationary phase and a gradient elution concerning acetonitrile. The 2 mm inner diameter of the chosen column results in a comparable small amount of required eluent. When compared with an optimized HPLC method using 3 µm particles in combination with a 3 mm column ID, saving is 73 % with respect to the eluent consumption and 68 % with respect to the analysis time. Accessorily, there is the possibility to use a shorter column if the separation of BBP and DBP is not of interest, because all other substances are separated with high resolution values. Shortening the column would lead to a shorter analysis time and therewith again to less usage of the mobile phase.

References

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4. Dr. Urs Hauri, Kantonales Laboratorium Basel Stadt. Duschgele in Weich-PVC-Verpackungen / Phthalate und deren Retention auf der Haut. 31.12.2008. www.kantonslabor-bs.ch/files/berichte/Duschgel08_2.pdf

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Physical properties of recommended column



BlueOrchid C18 use hydrophobic interactions for separation mechanism and offers an extended pH range for analysis of acidic, basic and neutral analytes in reversed phase mode. All BlueOrchid phases feature exceptional peak symmetry and resolution. Due to the narrow particle size distribution, the column back pressure of all BlueOrchid columns is lower than other high speed column materials on the market.

Stationary phase	BlueOrchid 1.8 C18
USP code	L1
Pore size	180 Å
Particle size	1.8 µm
Form	spherical
Surface area	180 m ² /g
% C	10
Endcapping	yes
Dimensions	100 x 2 mm
Order number	10BI181BOE

Recommended instrumentation



This application requires the PLATINblue binary high pressure gradient UHPLC system equipped with degasser, autosampler, column thermostat and PDA detector. Other configurations are also available. Please contact KNAUER to configure a system that is perfect for your needs.

Description	Order No.
PLATINblue HP system with PDA	A69420
PLATINblue Pump P-1, incl. 5ml pump head	
PLATINblue Pump-P1, incl. 5ml pump head and degasser	
HPG SmartMix 100	
PLATINblue Autosampler AS-1	
PLATINblue Column Thermostat T-1	
PLATINblue Detector PDA-1	
PDA flow cell (10 mm, 2 µl)	
ChromGate Software with PDA license	
PLATINblue UHPLC method converter	
PLATINblue stainless steel capillary kit	

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