KNAUER

Application Note

Fast analysis of water pollutant pesticides

Category	Environmental analysis
Matrix	-
Method	UHPLC
Keywords	Pesticides, herbicides, phenylurea pesticides, triazines, chloroacetanilide pesticides
Analytes	Atrazine, Chlorotoluron, Desethylatrazine, Isoproturon, Linuron, Metazachlor, Methabenzthiazuron, Metobromuron, Metolachlor, Metoxuron, Monolinuron, Simazine, Terbuthylazine
ID	**VEV0007N**, 10/12 rev.2

PLATIN blue

Summary

A very fast gradient method for the simultaneous determination of 13 pesticides which are known to be important water pollutants is presented in this application note. The high speed and reliability of this method applying the KNAUER PLATINblue UHPLC System make it well-suited for routine analysis. Reduction of analysis time to less than 8 minutes 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 used at a flow rate of 0.6 ml/min in combination with a UV detector.

Introduction

Pesticide residues in drinking water caused by the agricultural use of these substances in a significant rate are a serious problem today. In most of the cases these compounds act as herbicides, insecticides, nematicides, molluscicides and acaricides.[1] The adopted substances can consist of different chemical classes. In the majority of cases phenylureas, triazines, and chloroacetanilides are used. Many pesticides are known to have persistency and toxicological effects in the environment and to be potential endocrine disrupters.[1] As a conclusion they have to be regarded as environmental pollutants and their monitoring in ground and drinking water becomes inevitable.

According to the European Union directive on water quality (98/83/CE) the maximum concentration admissible for levels of pesticide residues in drinking and surface water is 0.10 µg/l for individual and 0.50 µg/l for the sum of pesticides.[2] It is obvious that the development of a reliable analysis method with high sensitivity and selectivity is important to comply with this directive. Additionally the established method has to be time and money saving so that it can easily be used in routine laboratories. Most pesticides are polar and thermally labile substances so that the classical analysis of environmental contaminants with gas chromatographic methods is not suitable. As a conclusion, HPLC and UHPLC are the methods of choice.[1]

In this application note a UHPLC method is presented that separates 13 important water pollutant pesticides in less than 8 minutes. The great advantage of employing a UHPLC method is saving of analysis time and money caused by the high speed and minimized eluent consumption.

Experimental sample preparation

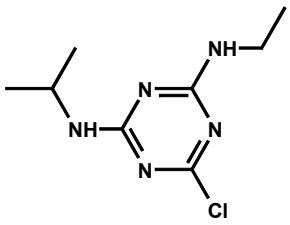
Controlling the pesticide concentrations is especially important in ground- and drinking water. Samples from drinking water can be collected, dechlorinated, preserved, shipped and stored as described by the EPA methods 532 or 535 for example. According to these methods, sample enrichment techniques like solid phase extraction or liquid-liquid-extraction must be used prior to the analysis.[3,4] This step is needed because the admissible level of individual pesticides according to the European Union directive on water quality is only 0.10 µg/l. Hence a water sample has to be preconcentrated to gain quantifiable results in this range.

Experimental preparation of standard solution

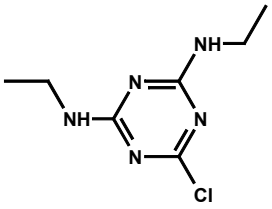
Standards were prepared by weighing out the exact masses for every pesticide compound and dissolving them in the mobile phase acetonitrile/water 20:80 (v/v). The concentrations of the stock solution for every component are shown in the following table.

Component	Concentration [mg/ml]
Metoxuron	1.0
Simazine	0.7
Methabenzthiazuron	1.0
Atrazine	0.7
Chlorotoluron	0.8
Monolinuron	1.1
Isoproturon	0.8
Metobromuron	1.2
Diuron	1.0
Metazachlor	1.2
Terbuthylazine	0.8
Linuron	1.0
Metolachlor	1.4

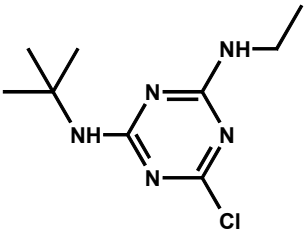
Pesticides from the Triazine class



Atrazine

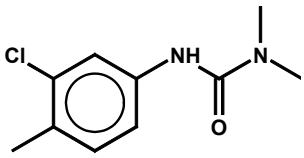


Simazine

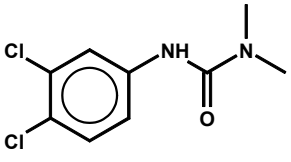


Terbuthylazine

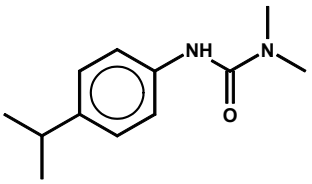
Phenylurea pesticides



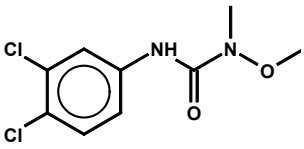
Chlorotoluron



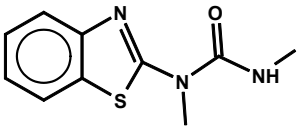
Diuron



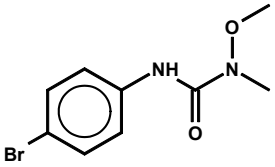
Isoproturon



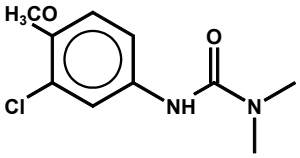
Linuron



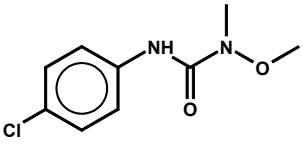
Methabenzthiazuron



Metobromuron

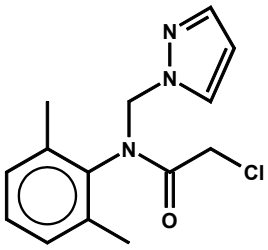


Metoxuron

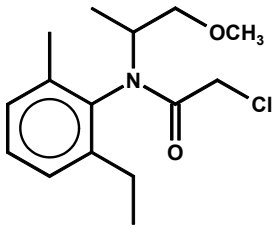


Monolinuron

Pesticides from the Chloroacetanilide class



Metazachlor



Metolachlor

Fig. 1

Chemical structures

Method parameters

Column	BlueOrchid 1.8 C18, 100 x 2 mm		
Eluent A	Water		
Eluent B	Acetonitrile		
Gradient	**Time [min]**	**% A**	**% B**
	0.00	95	5
	0.50	95	5
	6.00	55	45
	8.00	5	95
Flow rate	0.6 ml/min		
Injection volume	2 µl		
Column temperature	40 °C		
System pressure	approx. 520 bar		
Detection	UV at 215 nm (50 Hz, 0.05 s)		
Analysis time	7.5 min		

Results

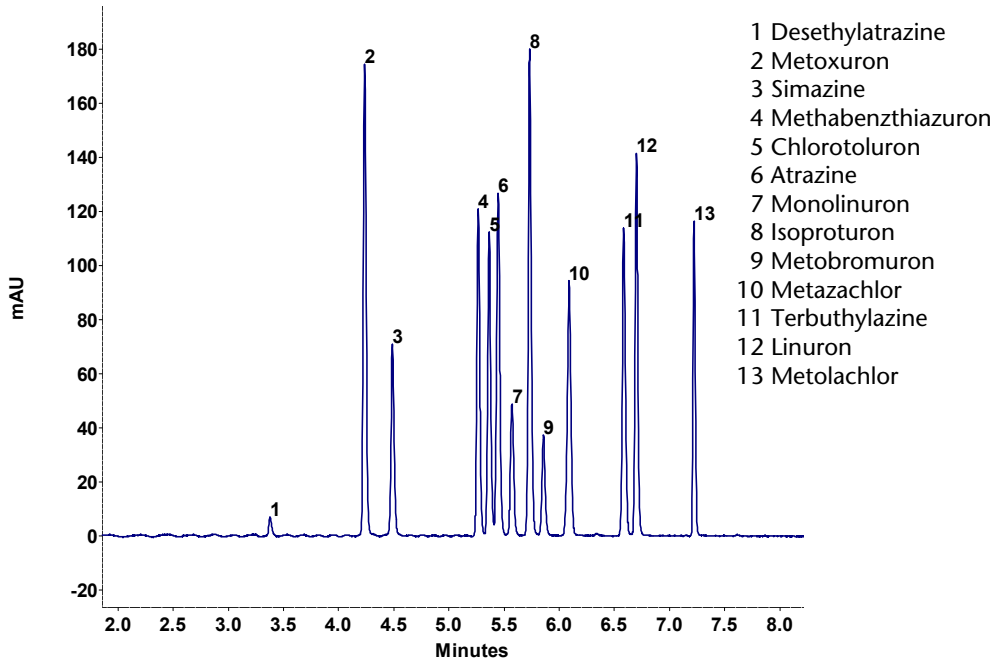


Fig. 2

Separation of the pesticide standard mix

Using the KNAUER PLATINblue UHPLC system and a BlueOrchid C18 1.8 µm column, 13 pesticides with significant occurrence in drinking water are successfully separated in less than 8 min (Fig. 2). All compounds are baseline separated with resolution values in the range of 1.6 for the critical pair chlorotoluron and atrazine to 16.1 for simazine and methabenzthiazuron.

Reliability of the UHPLC method is underlined by its retention time reproducibility in the range of 0.02 – 0.19 % RSD (n = 5) for all 13 pesticides.

Method performance

Resolution values	1.6 – 16.1
Retention time precision*	< 0.2 % RSD

*repeatability calculated over 5 replicate runs

Conclusion

This application note describes a very fast method for the determination of 13 pestizides with significant occurrence in drinking and ground water. The easy separation in less than 8 minutes becomes possible 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 78 % with respect to the eluent consumption and 70 % with respect to the analysis time.

References

1. J.M.F. Nogueira, Tom Sandra, Pat Sandra. Multiresidue screening of neutral pesticides in water samples by high performance liquid chromatography–electrospray mass spectrometry. Analytica Chimica Acta 505 (2004) 209–215.
2. EU Council, Directive on the Quality of Water Intended for Human Consumption, 98/83/EC, 1998.
3. Office of Research and Development, National Exposure Research Laboratory (NERL), US Environmental Protection Agency. Method 532. Determination of Phenylurea Compounds in Drinking Water by Solid Phase Extraction and High Performance Liquid Chromatography with UV Detection - Revision 1.0., June 2000.
4. Office of Research and Development, National Exposure Research Laboratory (NERL), US Environmental Protection Agency. Method 535: Measurement of Chloroacetanilide and Other Acetamide Herbicide Degradates in Drinking Water by Solid Phase Extraction and Liquid Chromatography/Tandem Mass Spectrometry (LC/MS/MS). Revision 1.1, issued in April 2005.

Physical properties of recommended column

BlueOrchid C18 uses 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's perfect for your needs.

Description	Order No.
PLATINblue UHPLC System	A69420
PLATINblue Pump P-1	
PLATINblue Pump P-1 with Degasser	
PLATINblue Autosampler AS-1	
PLATINblue Column Thermostat T-1 Basic	
PLATINblue Detector PDA-1	
PDA-1 flow cell (10 mm, 2 µl)	
PLATINblue modular eluent tray	
PLATINblue ChromGate Data system	
PLATINblue ChromGate PDA license	
PLATINblue stainless steel capillary kit	

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