

HPLC Analysis of Carbofuran, Methomyl, and Carbaryl by the JWVA* Method (* Japan Water Works Association)

The 2001 revision of the JWVA Method added tests for methomyl and carbaryl n-methylcarbamate pesticides to the existing test for carbofuran¹⁾. The HPLC analysis of carbofuran was previously described in Application News No. L260 and L272. This Application News introduces examples of the analysis

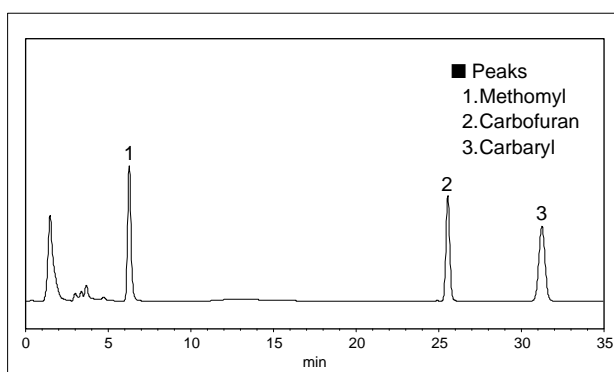
of three components – carbofuran, methomyl, and carbaryl – by post-column derivatization and fluorescence detection, and also by direct analysis with UV-Vis and fluorescence detections without derivatization.

■ Analysis of Carbofuran, Methomyl, and Carbaryl by Post-Column Derivatization

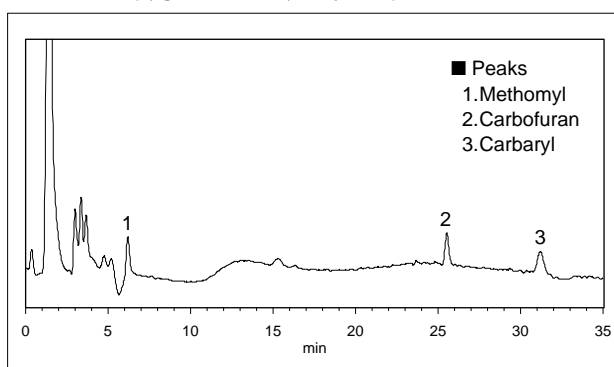
Post-column derivatization with o-phthalaldehyde (OPA) as the reaction reagent allows highly sensitive analysis of carbofuran, methomyl, and carbaryl. With this method, each pesticide component eluted in the column is first subjected on-line to hydrolysis with heating in an alkali solution, and then reacted with OPA for conversion to a strongly fluorescent substance.

Figs. 1 and 2 show the analysis results for a mixture of

carbofuran, methomyl, and carbaryl standards by the Shimadzu LC-VP Carbamate Analysis System, using the post-column derivatization method described above. The sample concentration was 5 µg/L per component for the analysis in Fig. 1, and 0.1 µg/L for the analysis in Fig. 2. The injected volume was 500 µL for each analysis. The feature of this system is its ability to measure samples with trace concentrations without concentration during pretreatment.



**Fig. 1 Chromatogram of a Mixture of Standards
(5 µg/L each, 500 µL injected)**



**Fig. 2 Chromatogram of a Mixture of Standards
(0.1 µg/L each, 500 µL injected)**

Table 1 Analytical Conditions

Column	:Shim-pack FC-ODS (75mmL.×4.6mmI.D.)
Mobile phase	:A→B Gradient elution A: Water, B: 2-Propanol
Flow Rate	:1.0mL/min
Temperature	:50°C (CTO-10ACVP)
Reagent 1	:0.05MNaOH
Flow Rate	:0.5mL/min
Temperature	:100°C(CRB-6A)
Reagent 2	:OPA solution (for carbamate analysis)
Flow Rate	:0.5mL/min
Temperature	:50°C(CTO-10ACVP)
Detection	:RF-10AXL Ex at 339nm, Em at 455nm

Table 2 Gradient Program

Initial B.Conc.	2%	
Time	Func	Value
6.00	B.Conc.	2
20.00	B.Conc.	15
32.00	B.Conc.	15
32.01	B.Conc.	2
44.00	STOP	

■ Repeatability of the Post-column Derivatization System

Repeatability testing was conducted on the system using the 0.1 μ g/L quantitation lower limit for each component prescribed in the JWWA method. Table 3 shows the results. Satisfactory repeatability was achieved, even at these extremely low concentrations.

Table 3 Peak Area Repeatability for Methomyl, Carbofuran, and Carbaryl

	Methomyl	Carbofran	Carbaryl
NO.1	8444	6713	7391
NO.2	9670	7151	9302
NO.3	8760	7853	8062
NO.4	8760	6935	8615
NO.5	9333	7352	8961
NO.6	8387	6892	9381
NO.7	8600	6809	7872
C.V.(%)	5.36	5.57	8.92

(0.1 μ g/L each, 500 μ L inj.)

Fig. 3 shows the analysis results for the injection of 500 μ L of a tap water sample spiked with standard solutions to 1 μ g/L concentration for each component. The chromatogram shows good resolution of bendiocarb and propoxur near the carbofuran peak.

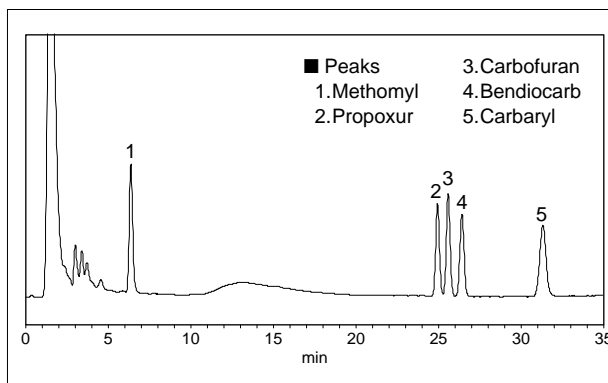


Fig. 3 Chromatogram of Tap Water (spiked 1 μ g/L each, 500 μ L injected)

■ Analysis of Carbofuran, Methomyl, and Carbaryl by UV-Vis and Fluorescence Detection

The fluorescence detector can detect carbofuran and carbaryl directly; Methomyl can be directly detected by the UV-Vis detector.

Fig. 4 shows the analysis results for the injection of 10 μ L of a tap water sample spiked with standard solutions to 200 μ g/L concentration for each component.

The repeatability results for the 200 μ g/L-concentration sample in Table 5 indicate a satisfactory result for each pesticide.

The JWWA method prescribes a 0.4 μ g/L quantitation

lower limit for this method. A 500x concentration in accordance with the method should easily clear this requirement.

Table 4 Analytical Conditions

Column	:Shim-pack VP-ODS (150mmL. \times 4.6mmI.D.)
Mobile phase	:Water/Acetonitrile = 7/3 (v/v)
Flow Rate	:1.0mL/min
Temperature	:40°C (CTO-10ACVP)
Detection	:SPD-10AVVP at 235nm RF-10AXL Ex at 279nm, Em at 307nm

Table 5 Peak Area Repeatability for Methomyl, Carbofuran, and Carbaryl

	Methomyl	Carbofran	Carbaryl
NO.1	6113	22069	57651
NO.2	6128	21999	57494
NO.3	6041	21728	57113
NO.4	6087	21201	58246
NO.5	6145	21185	59265
NO.6	6156	21226	58292
C.V.(%)	0.69	1.92	1.32

(200 μ g/L each, 10 μ L inj.)

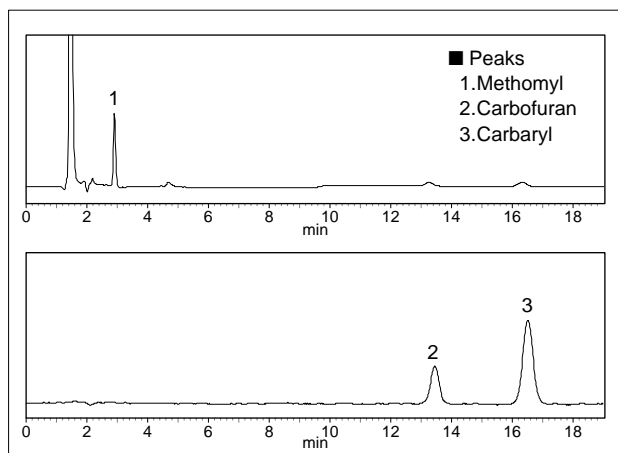


Fig. 4 Chromatogram of Tap Water (spiked 200 μ g/L each, 10 μ L injected)
Upper: SPD-10AVVP at 235 nm
Lower: RF-10AXL; Ex at 279 nm, Em at 307 nm

Reference

- 1) JWWA Test Method, 2001 Edition, Japan Water Works Association (2001)



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