

Comparing the Performance and Reliability of Waters Alliance HPLC Systems for Carbamate Analysis

Mark E. Benvenuti

Waters Corporation

Abstract

With the release of the 2013 Alliance HPLC System, this application note shows equivalency of the Waters Alliance System for Carbamate Analysis and 2013 Alliance HPLC System relative to linearity, precision, reproducibility, and limit of detection (LOD). The data shows that similar results are obtained on the legacy and 2013 Alliance HPLC systems for Carbamate Analysis.

Benefits

Waters Alliance System for Carbamate Analysis provides a complete system solution for the analysis of N-methylcarbamate and N-methylcarbamoyloxime pesticides in drinking water and a variety of environmental and food matrices.

- Carbamate analysis at sub-ppb levels
- Compatible with US EPA Method 531.2
- Baseline separation of analytes

Introduction

The Waters Alliance System for Carbamate Analysis has been offered as a system solution since 1998. With the release of the 2013 Alliance HPLC System, we wish to show equivalency of the two platforms relative to linearity, precision, reproducibility, and limit of detection (LOD).



Figure 1. 2013 Alliance HPLC System.

Experimental

LC conditions

| | |
|-------------------|---|
| System: | Alliance HPLC for Carbamate Analysis (both legacy and 2013 systems) |
| Run time: | 25.0 min |
| Column: | Waters Carbamate 3.9 x 150 mm, 4.0 µm at 30 °C |
| Mobile phase A: | Water |
| Mobile phase B: | Methanol |
| Mobile phase C: | Acetonitrile |
| Flow rate: | 1.5 mL/min |
| Injection volume: | 400 µL (1000 µL for 0.2- and 0.1-ppb levels) |
| Detection: | Fluorescence (Ex-339 nm, Em-445 nm) |
| Data management: | Empower 2 Software |

Gradient

| Time (min) | Flow rate (mL/min) | %A | %B | %C | Curve |
|---------------|--------------------------|----|----|----|-------|
| Initial | 1.5 | 88 | 12 | 0 | * |
| 5.3 | 1.5 | 88 | 12 | 0 | 1 |
| 5.4 | 1.5 | 68 | 16 | 16 | 5 |
| 14.0 | 1.5 | 68 | 16 | 16 | 3 |
| 16.1 | 1.5 | 50 | 25 | 25 | 7 |
| 20.0 | 1.5 | 50 | 25 | 25 | 6 |
| 22.0 | 1.5 | 88 | 12 | 0 | 5 |

The basic system, components, and experimental procedure are described in the Waters Alliance System for Carbamate Analysis Method Manual.¹

Standard and sample preparation

Two reference materials, M531M and M531-IS, were purchased from AccuStandard. These were diluted with preservation solution² to prepare the following levels: 100-, 75-, 50-, 25-, 10-, 5-, and 1-ppb. 0.2- and 0.1-ppb mixes were also prepared. These levels were also run as unknowns for the tests described in this study.

Linearity

The seven levels described above were injected in triplicate to construct a linear calibration curve.

Precision and reproducibility

The 75-, 25-, and 10-ppb levels were injected seven times as unknowns to determine precision and reproducibility for amount. The 21 injections (seven of each of the three levels) were used to determine precision and reproducibility for retention time.

Limit of detection

A 0.2-ppb carbamate mix was run three times as a standard, then seven times as an unknown to determine a limit of detection per 40 CFR pt 136 App, B³. A 0.1-ppb mix was also run for comparative purposes.

Performance evaluation

A Performance Evaluation Carbamate mix was purchased from ERA. The mix was prepared as directed, and quantified using both systems.

Results and Discussion

Figures 2 and 3 show overlaid chromatograms of the standards on the legacy and 2013 Alliance platforms, respectively. The linearity on both platforms is compared in Table 1. R^2 is greater than 0.999 for all compounds regardless of which platform was used. The reproducibility for both retention time and concentration was investigated for both platforms using seven injections of three different standard concentrations (10-, 25-, and 75-ppb) with the results shown in Tables 2 through 5. The relative standard deviation for the retention time for the 21 injections was less than 0.25% on both platforms, as shown in Table 2. Tables 3 through 5 show comparisons for the amount for seven injections each of the 75-, 25-, and 10-ppb carbamate mixes run as unknowns. The %RSD was less than 0.8% for the 75-ppb and 0.9% for the 25-ppb mixes using both the legacy and 2013 Alliance platforms. One exception to this is 1-Naphthol, which is a hydrolysis product of carbaryl⁴ and is, therefore, expected to show more variability. The %RSD was shown to be less than 1.5% for the 10-ppb mix on both platforms.

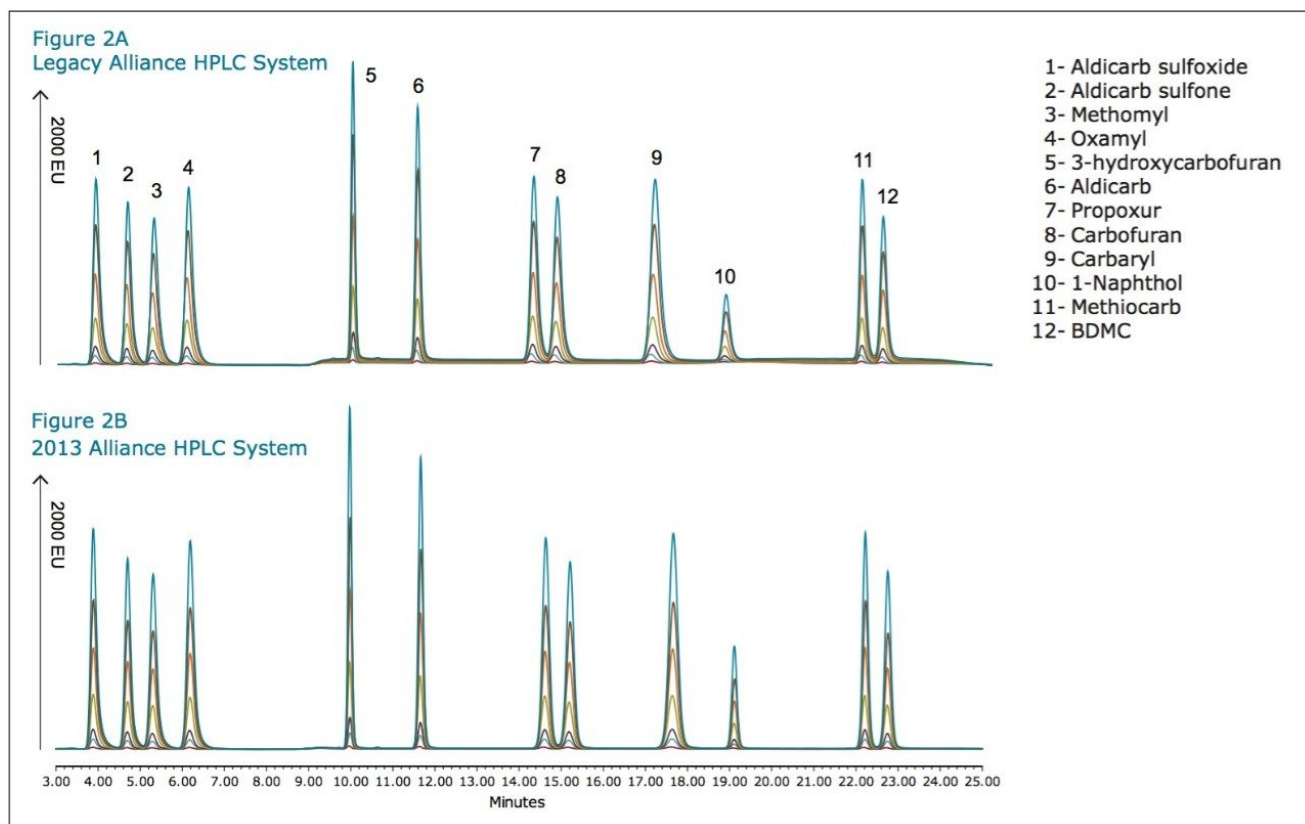


Figure 2. Chromatograms of 100-, 75-, 50-, 25-, 5-, and 1-ppb carbamate mixes using the legacy and 2013 Alliance HPLC systems.

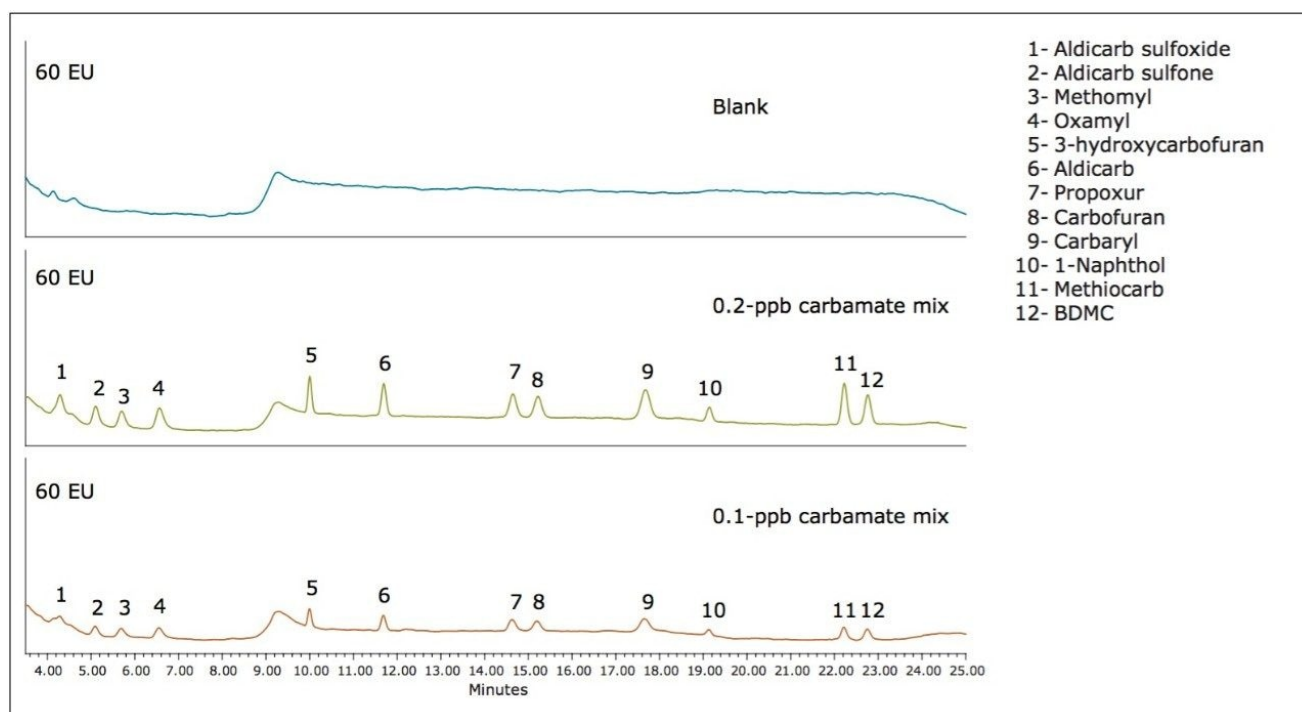


Figure 3. Chromatograms of low-level carbamate mixes on the legacy Alliance HPLC System, 1000- μ L injection.

| Linearity (R ²) | Legacy Alliance HPLC | 2013 Alliance HPLC |
|-----------------------------|----------------------|--------------------|
| Aldicarb sulfoxide | 0.9998 | 0.9998 |
| Aldicarb sulfone | 0.9998 | 0.9997 |
| Oxamyl | 0.9997 | 0.9998 |
| Methomyl | 0.9997 | 0.9997 |
| 3-Hydroxycarbofuran | 0.9998 | 0.9997 |
| Aldicarb | 0.9998 | 0.9998 |
| Propoxur | 0.9997 | 0.9998 |
| Carbofuran | 0.9997 | 0.9996 |
| Carbaryl | 0.9998 | 0.9996 |
| 1-Naphthol | 0.9998 | 0.9995 |
| Methiocarb | 0.9998 | 0.9996 |
| BDMC | 0.9999 | 0.9996 |

Table 1. Linearity for carbamate analytes.

| Reproducibility (RT) 21 injections | Legacy Alliance HPLC | | | 2013 Alliance HPLC | | |
|---|-----------------------------|----------------|-------------|---------------------------|----------------|-------------|
| Analyte | Mean RT | Std Dev | %RSD | Mean RT | Std Dev | %RSD |
| Aldicarb sulfoxide | 3.86 | 0.003 | 0.086 | 3.94 | 0.005 | 0.132 |
| Aldicarb sulfone | 4.59 | 0.008 | 0.174 | 4.65 | 0.008 | 0.170 |
| Oxamyl | 5.18 | 0.010 | 0.193 | 5.27 | 0.011 | 0.209 |
| Methomyl | 5.99 | 0.012 | 0.197 | 6.08 | 0.011 | 0.186 |
| 3-Hydroxycarbofuran | 9.94 | 0.009 | 0.089 | 9.93 | 0.007 | 0.072 |
| Aldicarb | 11.44 | 0.012 | 0.104 | 11.44 | 0.011 | 0.100 |
| Propoxur | 14.12 | 0.017 | 0.119 | 14.13 | 0.020 | 0.144 |
| Carbofuran | 14.65 | 0.018 | 0.125 | 14.67 | 0.021 | 0.146 |
| Carbaryl | 16.89 | 0.025 | 0.146 | 16.93 | 0.027 | 0.158 |
| 1-Naphthol | 18.63 | 0.021 | 0.114 | 18.62 | 0.018 | 0.097 |
| Methiocarb | 21.87 | 0.013 | 0.059 | 21.85 | 0.013 | 0.062 |
| BDMC | 22.36 | 0.013 | 0.059 | 22.33 | 0.015 | 0.065 |

Table 2. Reproducibility data for retention time for 21 injections, seven injections each, of 75-, 25-, and 10-ppb carbamate mixes using the legacy and 2013 Alliance HPLC systems.

| Amount 7 injections (75-ppb) | Legacy Alliance HPLC | | | 2013 Alliance HPLC | | |
|---|-----------------------------|----------------|-------------|---------------------------|----------------|-------------|
| Analyte | Mean RT | Std Dev | %RSD | Mean RT | Std Dev | %RSD |
| Aldicarb sulfoxide | 75.0 | 0.41 | 0.552 | 74.9 | 0.419 | 0.559 |
| Aldicarb sulfone | 75.3 | 0.46 | 0.610 | 75.2 | 0.367 | 0.488 |
| Oxamyl | 75.3 | 0.59 | 0.779 | 75.5 | 0.246 | 0.327 |
| Methomyl | 74.3 | 0.69 | 0.415 | 77.1 | 0.254 | 0.330 |
| 3-Hydroxycarbofuran | 75.9 | 0.32 | 0.415 | 74.7 | 0.318 | 0.427 |
| Aldicarb | 74.0 | 0.46 | 0.621 | 77.2 | 0.331 | 0.429 |
| Propoxur | 75.4 | 0.37 | 0.493 | 73.5 | 0.309 | 0.421 |
| Carbofuran | 77.0 | 0.25 | 0.329 | 76.3 | 0.303 | 0.398 |
| Carbaryl | 75.2 | 0.31 | 0.416 | 77.3 | 0.251 | 0.324 |
| 1-Naphthol | 70.0 | 0.92 | 1.321 | 88.3 | 0.289 | 0.327 |
| Methiocarb | 76.0 | 0.36 | 0.479 | 74.9 | 0.314 | 0.419 |
| BDMC | 76.6 | 0.44 | 0.572 | 76.2 | 0.353 | 0.464 |

Table 3. Reproducibility data for the amount of seven injections of the 75-ppb carbamate mix using the legacy and 2013 Alliance HPLC systems.

| Amount 7 injections (25-ppb) | Legacy Alliance HPLC | | | 2013 Alliance HPLC | | |
|---|-----------------------------|----------------|-------------|---------------------------|----------------|-------------|
| Analyte | Mean RT | Std Dev | %RSD | Mean RT | Std Dev | %RSD |
| Aldicarb sulfoxide | 25.4 | 0.099 | 0.391 | 24.8 | 0.133 | 0.539 |
| Aldicarb sulfone | 25.4 | 0.083 | 0.328 | 24.9 | 0.148 | 0.594 |
| Oxamyl | 25.6 | 0.108 | 0.422 | 25.1 | 0.076 | 0.303 |
| Methomyl | 25.3 | 0.106 | 0.420 | 25.6 | 0.122 | 0.477 |
| 3-Hydroxycarbofuran | 25.7 | 0.095 | 0.369 | 24.8 | 0.115 | 0.465 |
| Aldicarb | 24.9 | 0.124 | 0.499 | 25.6 | 0.158 | 0.619 |
| Propoxur | 25.2 | 0.130 | 0.515 | 24.4 | 0.104 | 0.425 |
| Carbofuran | 25.8 | 0.108 | 0.418 | 25.4 | 0.154 | 0.607 |
| Carbaryl | 25.1 | 0.100 | 0.399 | 25.7 | 0.108 | 0.420 |
| 1-Naphthol | 24.3 | 0.319 | 1.313 | 29.4 | 0.226 | 0.771 |
| Methiocarb | 25.4 | 0.113 | 0.445 | 24.7 | 0.220 | 0.889 |
| BDMC | 25.1 | 0.154 | 0.614 | 25.6 | 0.216 | 0.843 |

Table 4. Reproducibility data for amount for seven injections of the 25-ppb carbamate mix using the legacy and 2013 Alliance HPLC systems.

| Amount 7 injections (10-ppb) | Legacy Alliance HPLC | | | 2013 Alliance HPLC | | |
|---|-----------------------------|----------------|-------------|---------------------------|----------------|-------------|
| Analyte | Mean RT | Std Dev | %RSD | Mean RT | Std Dev | %RSD |
| Aldicarb sulfoxide | 10.1 | 0.034 | 0.338 | 9.7 | 0.062 | 0.638 |
| Aldicarb sulfone | 10.0 | 0.041 | 0.415 | 9.7 | 0.070 | 0.724 |
| Oxamyl | 10.2 | 0.051 | 0.501 | 9.8 | 0.067 | 0.678 |
| Methomyl | 10.0 | 0.049 | 0.490 | 9.9 | 0.094 | 0.939 |
| 3-Hydroxycarbofuran | 10.2 | 0.041 | 0.405 | 9.7 | 0.077 | 0.794 |
| Aldicarb | 9.8 | 0.059 | 0.602 | 10.0 | 0.106 | 1.055 |
| Propoxur | 10.0 | 0.057 | 0.578 | 9.6 | 0.076 | 0.793 |
| Carbofuran | 10.2 | 0.067 | 0.655 | 10.0 | 0.100 | 1.005 |
| Carbaryl | 10.0 | 0.042 | 0.419 | 10.0 | 0.041 | 0.410 |
| 1-Naphthol | 10.2 | 0.099 | 0.967 | 11.6 | 0.146 | 1.262 |
| Methiocarb | 10.1 | 0.071 | 0.698 | 9.5 | 0.088 | 0.924 |
| BDMC | 10.3 | 0.142 | 1.368 | 9.5 | 0.098 | 1.032 |

Table 5. Reproducibility data for amount for seven injections of the 10-ppb carbamate mix on the legacy and 2013 Alliance HPLC systems.

To investigate the detection of low levels of the pesticides, a blank, 0.2-ppb, and 0.1-ppb carbamate mix was injected with the resulting chromatograms shown in Figures 3 and 4. The calculated limits of detection, shown in Table 6, were 0.1-ppb or less for both systems with the exception of 1-Naphthol which, as previously mentioned, is a degradation product.

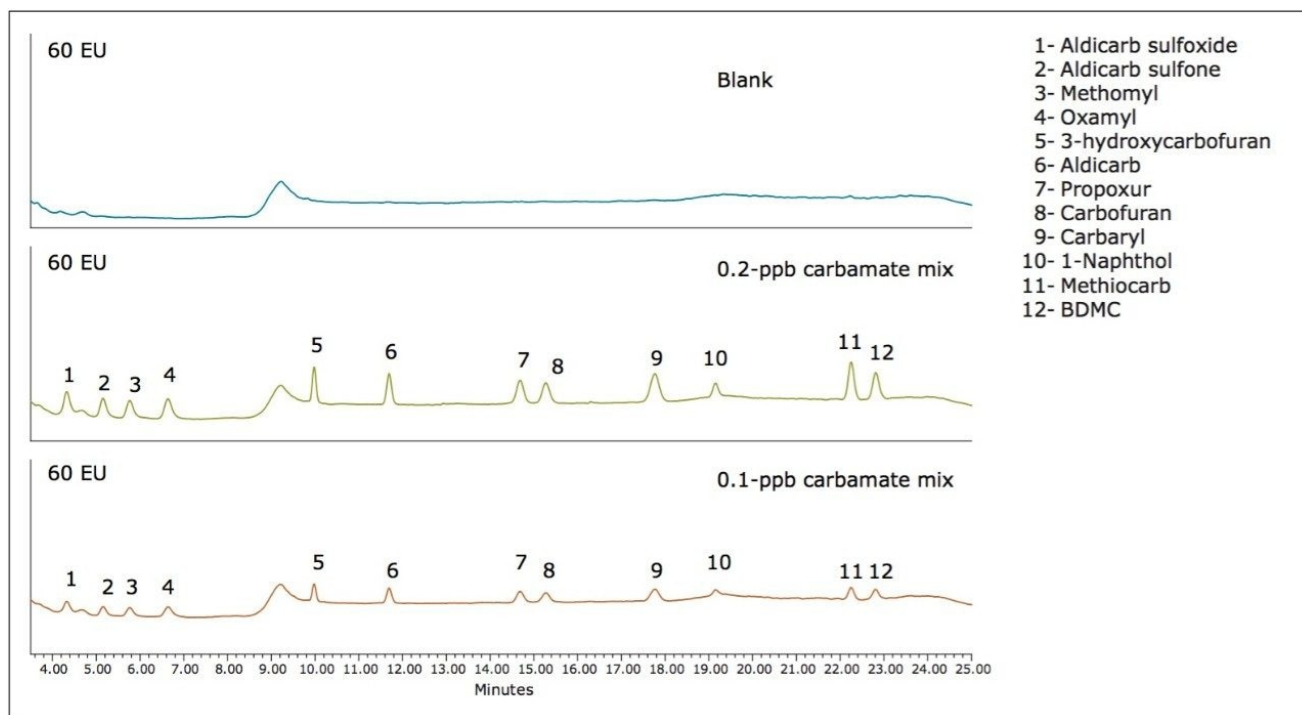


Figure 4. Chromatograms of low-level carbamate mixes on the 2013 Alliance HPLC System, 1000- μ L injection.

| Limit of detection | Legacy Alliance HPLC | 2013 Alliance HPLC |
|---------------------|----------------------|--------------------|
| Aldicarb sulfoxide | 0.05 | 0.03 |
| Aldicarb sulfone | 0.06 | 0.03 |
| Oxamyl | 0.10 | 0.04 |
| Methomyl | 0.06 | 0.07 |
| 3-Hydroxycarbofuran | 0.09 | 0.02 |
| Aldicarb | 0.02 | 0.03 |
| Propoxur | 0.04 | 0.03 |
| Carbofuran | 0.07 | 0.10 |
| Carbaryl | 0.09 | 0.05 |
| 1-Naphthol | 0.19 | 0.36 |
| Methiocarb | 0.04 | 0.04 |
| BDMC | 0.07 | 0.10 |

Table 6. MDL data for seven injections of a 0.2-ppb mix calculated per 40 CFR pt 136 App B (ppb).

To test the two systems using a blind sample, the ERA performance evaluation sample was used. For both the previous and current systems, the calculated amounts were found to be within the acceptable QC performance limits and within 1-ppb of each other, as shown in Table 7.

| ERA Certified Mix Lot #5180-707 | Cert value | QC acceptable range | Legacy Alliance HPLC | 2013 Alliance HPLC |
|--|-------------------|--------------------------------|---------------------------------|-------------------------------|
| Aldicarb sulfoxide | 16.2 | 11.9-21.6 | 13.7 | 13.3 |
| Aldicarb sulfone | 19.0 | 14.2-23.4 | 17.9 | 18.0 |
| Oxamyl (Vydate) | 70.9 | 52.5-85.8 | 59.8 | 58.9 |
| Methomyl | 53.7 | 41.1-67.1 | 46.9 | 46.2 |
| 3-Hydroxycarbofuran | 45.6 | 34.8-57.0 | 39.6 | 38.9 |
| Aldicarb | 30.1 | 21.9-37.9 | 26.4 | 26.0 |
| Propoxur (Baygon) | 42.1 | 32.5-51.8 | 36.7 | 36.2 |
| Carbofuran | 39.5 | 27.2-50.2 | 35.1 | 34.3 |
| Carbaryl | 24.4 | 16.9-29.8 | 20.3 | 20.0 |
| Methiocarb | 82.8 | 61.4-97.7 | 69.8 | 69.1 |

Table 7. Analysis of ERA QC carbamate mix units are ppb.

Conclusion

The data show that similar results are obtained on the legacy and 2013 Alliance HPLC systems for Carbamate Analysis, yielding the following benefits:

- Conformance to EPA Method 531.2
- Baseline separation of analytes
- Run time of 25 minutes
- Provides a complete Waters solution for carbamate analysis

References

1. Waters Alliance System for Carbamate Analysis Method Guide, p/n 71500017101 Rev D.

2. *Ibid.* 3-11.
3. US EPA 40 CFR pt 136 App, B. 565-566.
4. de Bertrand, et al. Photodegradation of the Carbamate Pesticides Aldicarb, Carbaryl and Carbofuran in Water. *Analytica Chimica Acta*. 1991; 254:235-244.

Featured Products

Alliance HPLC System <<https://www.waters.com/534293>>

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