TECHNICAL NOTE:

EVALUATION OF HDPE WATER SAMPLE BOTTLES AND PVC SAMPLER TUBING USED IN HERBICIDE DISSIPATION STUDIES

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ABSTRACT. *The recovery of six herbicides (triclopyr, triclopyr ester, sulfometuron methyl, metsulfuron methyl, imazapyr, and hexazinone) was evaluated in two stream water samples, one from Weogufka Creek in the Alabama Piedmont and one from a stagnant stream in the Escambia Experimental Forest near Florida. Simulated field study conditions were used to evaluate collection, ambient (pre-retrieval) field storage, and freezer storage phases of stream water sampling. Method detection levels were developed for each herbicide in each water matrix. None of the herbicides degraded significantly in either stream water after 24 days at 22°C to 25°C or after 12 months of freezer storage below -15°C. None of the herbicides tested appeared to leach from the automatic sampler tubing into subsequently collected samples. However, slight carryover of about 0.8% of the spiking level was observed in the first post-spike blanks collected for all herbicides. This appeared to be the result of physical adhesion of water droplets on tubing walls; no detectable carryover was observed in subsequent blanks. These results support the use of HDPE sample containers and flexible PVC automatic sampler tubing in environmental fate studies of herbicides.*

Keywords. *Automatic water samplers, Environmental fate studies, Forest watersheds, HDPE sample bottles, Herbicides, High-performance liquid chromatography, PVC tubing.*

In 40 CFR, Part 158, Subpart N, the EPA requires pesticide manufacturers to submit offsite movement and persistence data for every pesticide registered for use in the U.S. and its territories. These requirements must be satisfied by field studies under actual use conditions, called environmental fate studies. Herbicides used in forest management constitute a special class of pesticides, requiring that these studies be performed on isolated and often mountainous forest watersheds, far from paved access roads. Over the past 20 years, forestry research teams have conducted studies of the dissipation and fate of herbicides in forests of the southeastern U.S. (Bush et al., 1988, 1990; Miller and Bace, 1980; Neary, 1983, 1985; Bouchard et al., 1985; Neary et al., 1983, 1985, 1986, 1993; Neary and Michael, 1989, 1996; Michael and Neary, 1991, 1993; Michael et al., 1989, 1994, 1999). Similar studies have also been conducted in Arizona (Davis and Ingebo, 1973; Johnsen, 1980), West Virginia (McKellar et al., 1982; Lavy et al., 1989), Oregon (Norris and Montgomery, 1975; Norris, 1981, Newton et al., 1984), Washington State (Rashin and Graber, 1993), British Columbia (Wan, 1987), Ontario (Thompson et al., 1991), and Quebec (Legris et al., 1985; Legris, 1987, 1988; Legris and Couture, 1989).

The remoteness of suitable forested sites necessitates around-the-clock, unattended collection of stream runoff samples using automatic sampling devices. These devices depend on the use of high-density polyethylene (HDPE) bottles for sample collection and flexible polyvinyl chloride (PVC) tubing for aspiration of stream water samples into these bottles. During automatic sample collection cycles, traces of herbicides might potentially adsorb onto the autosampler tubing walls and contaminate subsequent samples. Sampling is usually most intensive during herbicide application and the first three or four substantial rainfalls thereafter, when most offsite movement has been found to occur. Sampling is less frequent between storms, when herbicide runoff returns to baseline levels, below minimum detection limits. Sample bottles are capped and transferred to freezers as rapidly as possible after they are collected, but study sites are often a day's journey from laboratories where samples can be stored and analyzed. Under typical field conditions, storm samples may remain uncapped in HDPE sample bottles inside samplers for up to 8 h before they can be retrieved, capped, and stored in freezers for transport. Non-storm samples can remain at ambient temperatures for up to 24 days, subject to evaporation and microbial growth. All samples are stored frozen until they are thawed just before analysis. All frozen samples are analyzed as soon as possible, but some may remain stored for up to one year. Freezer storage stability samples are commonly run alongside field samples for quality control. This is traditionally done by spiking analytically pure standards of the herbicide active ingredient(s) into distilled water, storing them in sample bottles in deep freezers, and analyzing them periodically.

Each of the three pre-analysis protocols described above is a necessary phase of every forestry herbicide environmental fate study. Each presents opportunities for systematic error that might bias the overall results. The purpose of this study was to examine each of these phases in isolation to determine its contribution to overall systematic error. The tests

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Submitted for review in May 2008 as manuscript number SW 7500; approved for publication by the Soil & Water Division of ASABE in September 2008.

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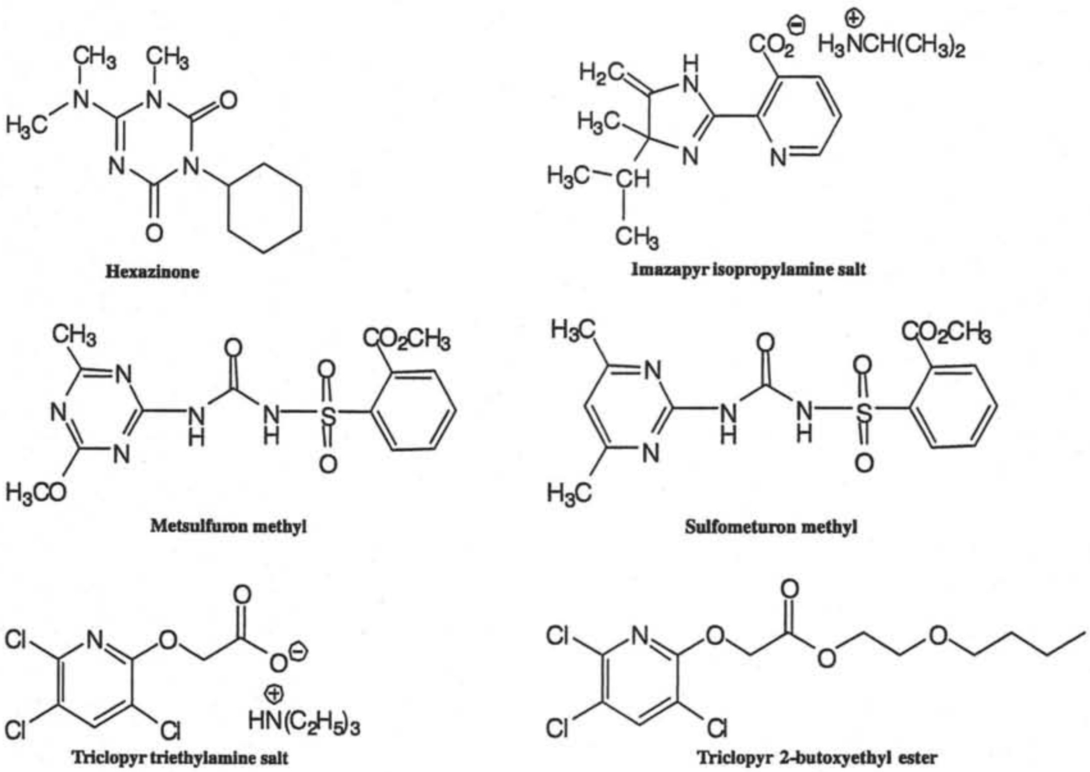

Figure 1. Chemical structures of the herbicides used in this study.

described here were designed to simulate the conditions experienced by real stream water samples during each phase and to measure their effects, if any, on herbicide recoveries. To our knowledge, this is the first such study to be performed independently of a specific environmental fate study, and the first to compare the behavior of several classes of herbicides under identical simulated field conditions.

In the freezer stability study, six commercial herbicide formulations were spiked into two different stream water samples, and then thawed and analyzed at intervals of up to one year. In the ambient temperature stability study, the herbicide-spiked stream waters were placed in uncapped sample bottles and left in covered auto-sampler carousels at 22°C to 25°C for up to six days, and then analyzed. In the tubing adsorbtion and leaching study, an automatic water sampler was used to draw herbicide-spiked stream water, followed by blank stream water, through known lengths of standard suction and peristaltic pump tubing. The blank samples were then analyzed to detect any carryover of herbicide.

MATERIALS AND METHODS

Six herbicides widely used in forestry were chosen for testing. These were: imazapyr isopropylamine salt (Arsenal AC), triclopyr triethylamine salt (Garlon 3A), hexazinone (Velpar L), metsulfuron methyl (Escort), sulfometuron methyl (Oust), and triclopyr ester (Garlon 4). Their chemical structures are shown in figure 1.

These herbicides were selected because they span a broad range of aqueous solubility and hydrophilic or hydrophobic character. Their relevant chemical properties are listed in table 1. These include octanol-water partition coefficient(s) (Kow), acid dissociation constant(s) (pKa), and aqueous solubilities.

Table 1. Relevant chemical properties of the herbicides in this study.

Herbicide	Log(Kow)	pKa @ 25°C	Solubility in H_2O @ 25°C
Imazapyr acid	1.3	1.9, 3.6	11.3×10^3 mg L^{-1}
Hexazinone	1.5	Not ionized	330 mg L^{-1}
Metsulfuron methyl	0.014 (pH 7) 1.00 (pH 5)	3.3	2.8×10^3 mg L^{-1} (pH 7) 548 mg L^{-1} (pH 5)
Sulfometuron methyl	0.31 (pH 7) 15 (pH 5)	5.7	300 mg L^{-1} (pH 7) 10 mg L^{-1} (pH 5)
Triclopyr acid	0.36 (pH 5) 2.64 (pH 7)	2.7	440 mg L^{-1} (pH 7)
Triclopyr BE ester	4.1	Not ionized	23 mg L^{-1}

Triclopyr triethylamine salt is water-miscible, but neutralizes to the insoluble free acid below pH 3.0. Imazapyr is an imidazole base as well as an acid and remains partially ionized and water-soluble at all pH levels.

HERBICIDE ANALYTICAL STANDARDS

Pure analytical standards of triclopyr and triclopyr 2-butoxyethylester were supplied by Dow AgroSciences (Midland Mich.). Certified standard imazapyr was donated by BASF Agricultural Products Group (Research Triangle Park, N.C.). Research-grade hexazinone was supplied by Dupont Agricultural Products (Wilmington, Del.). Analytically pure sulfometuron methyl and metsulfuron methyl were purchased from Supelco, Inc. (Bellefonte, Pa.).

HPLC OPERATING CONDITIONS

All herbicide residues were analyzed by reversed-phase HPLC on a Finnigan SpectraSYSTEM composed of a P4000

Table 2. Reversed-phase HPLC mobile phase compositions and detection wavelengths.

Herbicide	Mobile Phase Composition	UV Detection Wavelength
Imazapyr	20/80 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	240 nm
Triclopyr	45/55 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	232 nm
Triclopyr ester	60/40 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	232 nm
Sulfometuron methyl	45/55 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	230 nm
Metsulfuron methyl	30/70 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	224 nm
Hexazinone	30/70 (v/v) acetonitrile/water (pH 2.0 with H_3PO_4)	248 nm

programmable quaternary gradient pump, an AS3000 variable-loop auto-sampler, a UV1000 programmable ultraviolet absorbance detector, and a Chrom Jet SP4400 integrator (Thermo Electron Corp., San Jose, Cal.). The column used for all separations was a Zorbax Stablebond SB-C8 analytical column (5 µm, 150 × 4.6 mm id) with a Zorbax SB-C8 guard cartridge (12.5 × 4.6 mm id) (MAC-MOD Analytical, Chadds Ford, Pa.). The column was kept at 40°C by a Shimadzu CTO-6A forced-air column oven (Shimadzu, Kyoto, Japan). All mobile phases were isocratic mixtures of acetonitrile (Burdick and Jackson), high-purity water (Milli-Q System, Millipore, Bedford, Mass.), and reagent-grade phosphoric acid (Mallinckrodt, Phillipsburg, N.J.). Mobile phases and UV detection wavelengths were optimized for each herbicide and are shown in table 2. All mobile phases were pumped at a rate of 1.0 mL min^{-1}.

STREAM WATER SAMPLE COLLECTION

Samples of stream water were collected in 18.9 L polyethylene buckets from each of two sites. The buckets were purchased new, thoroughly washed with soap and water, and rinsed with deionized water prior to use. The stream water samples were stored in a cold room at 5°C until needed. One sample was taken from Weogufka Creek, a freely flowing stream on a piedmont loblolly pine plantation in Coosa County, Alabama (pH 6.5). This sample was clear, permitting analysis by direct-injection on reversed-phase HPLC. The other sample was from a stagnant stream in the Escambia Experimental Forest in southern Alabama (pH 5.1), a coastal plain site managed for longleaf pine. This sample was visibly amber colored from dissolved organic matter and contained suspended sediment. It required cleanup prior to analysis by HPLC with ultraviolet absorbance detection. These samples were chosen to represent the diversity of forested stream sites in the southeastern U.S.

SAMPLE CLEANUP FOR WEOGUFKA CREEK WATER

The sample bottles were capped tightly and agitated for 1 min to ensure uniformity. Two milliliters of each water sample was filtered through a Millipore Millex-HN 13 mm filter unit (Millipore Corp., Bedford, Mass.) into a 12 × 32 mm auto-sampler vial (Sun SRI, Wilmington, N.C.). Aliquots of 100 µL were directly injected on the HPLC system.

Hexazinone co-eluted with an interfering substance in the Weogufka Creek water, making HPLC quantitation unreliable. This interference could be removed using the solid-phase extraction procedure described below, but omitting the lead precipitation.

Triclopyr ester hydrolyzed rapidly in water and could only be detected as a mixture with hydrolyzed triclopyr. Being highly nonpolar and hydrophobic, triclopyr ester also adsorbed onto suspended sediment particles and the walls of sample bottles. This caused loss of the adsorbed fractions during the filtration step. It was found that triclopyr ester could be hydrolyzed fully to soluble triclopyr salt by adjusting the water sample to pH 10. This was accomplished by adding 200 µL of 1.0 M KOH to each sample bottle and allowing them to stand at room temperature for 24 h.

All the above treatments incorporated filtration to remove suspended sediments prior to analysis.

SAMPLE CLEANUP FOR ESCAMBIA STREAM WATER

The sample bottles were capped tightly and agitated for 1 min to mix uniformly. Then 100 mL of water from each sample bottle was decanted into a 250 mL beaker, and 200 µL of 1 M lead diacetate (J. T. Baker, Phillipsburg, Pa.) was added with stirring. The solutions were left to stand for 1 h to allow precipitates to coagulate and settle. Each sample was suction filtered through a 5.5 cm Whatman GF/B filter (Whatman, Inc., Clifton, N.J.). The filtrates were adjusted to pH 2.0 with 1 N hydrochloric acid (Mallinckrodt Chemical Company, Paris, Ky.) and subjected to solid-phase extraction (SPE) using BakerBond Octadecyl (C18) 1000 mg SPE columns (J. T. Baker). These columns gave recoveries of 91% to 99% for all the herbicides in this study except imazapyr, which yielded only 20% recovery.

SPE columns can differ markedly in retention from maker to maker and from batch to batch due to variations in the base silica and bonded phase coverage. Surface area and morphology, trace metal impurities, and exposed silanol sites all affect retention. Waters SepPak Environmental C_{18} 1000 mg SPE columns (Waters Corporation, Milford, Mass.) were found to improve the retention and recovery of imazapyr to 90%.

The SPE columns were mounted on a Supelco 12-place vacuum manifold (Supelco, Bellefonte, Pa.). Each SPE column was preconditioned with 5 mL of methanol (Burdick and Jackson, Muskegon, Mich.) followed by 10 mL of high-purity water from a Millipore Milli-Q Plus system acidified to pH 2.0 with 1.0 M HCl. The samples were passed through the columns at a rate of 4 mL min^{-1} or less. The columns were post-washed with another 10 mL of acidified water, and dried for 5 min by drawing air through them under vacuum. The columns were eluted with 5 mL of methanol, shaken to mix, and transferred to 12 × 32 mm auto-sampler vials. Aliquots of 10 µL were injected onto the HPLC system.

Both cleanup steps involved filtration, which removed any suspended sediment in the samples.

MDL DETERMINATIONS

Prior to sample analysis, the analytical methods were evaluated to determine the method detection limits (MDL), quantitation limits (QL), and reporting limits (RL) for each herbicide in each water matrix. Chromatographers have traditionally defined the MDL as 3 × the signal-to-noise ratio of the HPLC detector. However, in 40 CFR, Part 136, Appendix B, the EPA has defined MDL as the standard deviation for seven spikes × 3.14 (Student's t value for $n = 7$). The spiking levels used for MDL determination must be less than ten

Table 3. Detection, quantitation, and reporting levels of herbicides in stream water samples using reversed-phase HPLC (mean values; $n = 4$).

Herbicide	Weogufka Creek Water[a]		Escambia Stream Water[b]	
	MDL ($\mu g\ L^{-1}$)	QL&RL ($\mu g\ L^{-1}$)	MDL ($\mu g\ L^{-1}$)	QL&RL ($\mu g\ L^{-1}$)
Sulfometuron methyl	0.4	1.1	0.8	2.4
Triclopyr	0.9	2.8	0.3	0.8
Imazapyr	0.4	1.1	0.6	1.9
Metsulfuron methyl	0.5	1.4	0.3	1.0
Triclopyr BE ester (as acid)	0.9	2.8	0.5	1.7
Hexazinone	0.3[c]	0.9[c]	0.4	1.3

[a] Analyzed by direct aqueous injection.

[b] Analyzed by lead precipitation, filtration, and solid-phase extraction.

[c] Analyzed by acidification to pH 2.0 followed by solid-phase extraction.

times the MDL, so repeat analyses are often necessary. This gives a result similar to the 3 × signal-to-noise ratio definition and applies to peak area as well as peak height. The EPA defines the QL as ten times the standard deviation and the RL as greater than or equal to the QL. The target spiking level chosen for each of these studies was ten times the RL or 100 $\mu g\ L^{-1}$, whichever was greater. The actual spiking level measured at baseline (day 0) could vary from the target due to the titer of the herbicide active ingredient and the recovery level of the analytical method. MDL studies using the direct sample injection method were performed in the Weogufka Creek water matrix. MDL studies for the Escambia water matrix were performed using the SPE cleanup procedure with lead diacetate precipitation. The results are given in table 3.

Ambient Stability Study Protocol

Study analyses were performed on three sets of water samples, each consisting of four spikes and four blanks. Sufficient stream water was transferred to a 50 L carboy to make twenty-four 800 mL samples. Twenty-four clean ISCO 1 L HDPE sector bottles were weighed to the nearest ±0.01 g. Water (800 mL) was transferred into each bottle, and then the bottle was weighed again to give the initial mass of the sample. Suspended sediment was not measured, but settling occurred in the blending tank, and sediment was not visible in the test samples of either stream water. Twelve bottles were spiked with an aqueous herbicide formulation to give approximately 100 $\mu g\ L^{-1}$ active ingredient. The other twelve were left blank. This spiking level was chosen because it exceeds the peak runoff levels that we have observed in our field studies, and because it is well within the linear range for ultraviolet absorbance detection of all the herbicides studied. The samples that were spiked with sulfometuron methyl and metsulfuron methyl were first buffered with 1.0 mL of 0.1 M (pH 7.0) potassium phosphate to retard hydrolysis. The samples spiked with triclopyr ester were dosed with 200 μL of 1 M KOH to promote hydrolysis (discussed above).

Four spikes and four blanks were analyzed at once to determine baseline concentration. The remaining blanks and spikes were placed uncapped in an ISCO carousel at ambient temperature with its lid closed to simulate field conditions. The remaining sets of four blanks and four spikes each were analyzed after 2 days (Weogufka Creek), 6 days (Escambia), and 24 days of ambient storage (both water samples). The stored samples were reweighed to measure evaporation loss and analyzed at once by reversed-phase HPLC. The blanks and spikes were each averaged. All sample injections were duplicates and were averaged. Every four samples (eight injections) were bracketed by duplicate injections of external standards of 100 $\mu g\ L^{-1}$ active ingredient in deionized water. Chromatograms were quantified by electronic integration of peak areas. The stability at each storage time was calculated using equation 1:

$$\text{Percent remaining} = [(S_t - B_t)/(S_0 - B_0)]\cdot(V_0/V_t)\cdot 100\% \qquad (1)$$

where

S_t = average spike recovery in ng mL^{-1} at time t ($n = 4$)
B_t = average blank background interference in ng mL^{-1} at time t ($n = 4$)
S_0 = average initial spike recovery (ng mL^{-1}; $n = 4$)
B_0 = average initial blank background interference (ng mL^{-1}; $n = 4$)
V_0 = initial sample volume (mL)
V_t = final sample volume (mL) at time t.

Percent recovery vs. storage time was calculated for each herbicide in each stream water sample.

Freezer Stability Study Protocol

For the freezer stability study, each of the six herbicides was tested in the two stream waters from the Alabama Piedmont and Coastal Plain. Analogously to the ambient stability study, analyses were performed on four sets of water samples, each consisting of four spikes and four blanks. Sufficient water was transferred to the 50 L carboy to make thirty-two 800 mL samples, half of which were spiked with herbicide. All 32 samples were placed in chest freezers for storage at or below -15°C. One set of four spikes and four blanks was thawed after overnight freezing and analyzed to determine baseline concentrations. The remaining blanks and spikes were thawed and analyzed in quadruplicate sets after 3, 6, and 12 months of storage. The thawed samples were reweighed to determine evaporation loss during storage, shaken for 1 min to homogenize, and analyzed by HPLC. Percent remaining versus freezer storage time was calculated in the same manner as was done for the ambient stability study.

Auto-Sampler Tubing Adsorption and Leaching Study Protocol

Study analyses were performed on four sequences of three spikes followed by three blanks. An ISCO model 3700 automatic sampler was fitted with 9.1 m of ISCO 0.953 cm ID PVC suction tubing and 103.8 cm of 0.953 cm ID Silastic medical-grade silicone rubber pump tubing (ISCO, Inc., Environmental Division, Lincoln, Neb.). The sampler pump was elevated above the end of the suction tubing to give 1.5 to 2.5 m of head. The flexible PVC suction line tubing was supported on lengths of rigid PVC pipe to eliminate loops and low points, which could trap water. Single auto-sampler draw and back-flush cycles were used for each collection. Each cycle included an initial blowout to clear the tubing, followed by an 800 mL collection and final back flush. Separate tests were performed for each of the six herbicides in both stream-water types, for a total of 12 tests.

For each test, 12 L of stream water in an 18.9 L bucket was spiked with a sufficient volume of herbicide in tap water to give a final concentration of approximately 100 $\mu g\ L^{-1}$ active ingredient. A second bucket was filled with 14 L of unspiked stream water. An initial set of four blank samples was drawn from the unspiked bucket to flush the tubing and establish the background level. These samples were removed from the carousel and replaced with fresh bottles. The tubing was removed from the unspiked bucket and transferred to the spiked water bucket. A sequence of three spiked samples was drawn, followed by three blank samples, alternating the end of the tubing between the spiked and unspiked buckets after every three samples. This gave four repetitions of three spikes followed by three blanks. The overall sequence was four preliminary blanks followed by: 3 Spk → 3 Blk → 3 Spk → 3 Blk → 3 Spk → 3 Blk → 3 Spk → 3 Blk. The end of the tubing was shaken between transfers to minimize physical carryover of adhering drops. All samples were analyzed by HPLC the following day.

The percent carryover was calculated for each of the three successive post-spike blanks for each herbicide-stream water combination using equation 2:

$$C_i = 100\%\cdot(B_i - B_b)/S_i \tag{2}$$

where

S_i = mean (n = 4) recoveries in each of the sequential pre-blank spikes (ng mL^{-1}; i = 1-3)
B_i = mean of carryover levels (ng mL^{-1}) in each of the sequential post-spike blanks (i = 1-3)
B_b = mean of background interference levels (ng mL^{-1}) in the initial pre-spike blanks (n = 4).

Statistical Methods

Mean herbicide recoveries from stream waters after each sampling interval (n = 4 in all cases) were evaluated against their day 0 controls using the two-tailed paired Student's t-test. Differences were considered significant at the $P < 0.05$ level. Standard deviations and t-tests were calculated using the STDEV and TTEST functions in Microsoft Excel.

Results and Discussion

Ambient Stability of Herbicides in Stream Water Samples

The mean percentage recoveries of all six herbicides from stream water samples at day 2, day 6, and day 24 relative to day 0 are given in table 4. Most of the mean recoveries tested significant because the HPLC analyses were highly repeatable. Mean recoveries were well within normal variation observed in field studies. The overall means across the diverse range of herbicides tested reflect the viability of remote stream water sampling using automatic samplers.

Table 4. Recovery of herbicides from stream water samples following ambient temperature standing in HDPE automatic sampler bottles (mean values; n = 4).[a]

	Weogufka Creek Water		Escambia Stream Water	
Herbicide	Day 2	Day 24	Day 6	Day 24
Imazapyr	104.6 ±0.6*	93.4 ±2.0*	98.3 ±0.9*	99.3 ±0.5
Hexazinone	99.4 ±1.7	96.1 ±1.9*	100.1 ±0.5	100.0 ±0.5
Metsulfuron methyl	101.7 ±0.4*	100.0 ±0.8	100.0 ±1.1	97.1 ±0.4*
Sulfometuron methyl	97.7 ±0.8	91.1 ±0.9*	94.8 ±1.9*	84.3 ±3.9*
Triclopyr	95.9 ±0.8*	100.8 ±0.7	99.8 ±0.6	100.2 ±1.3
Triclopyr BE ester	103.9 ±3.6	106.1 ±0.7	100.1 ±1.5	100.5 ±2.6
Mean (all herbicides)	100.5 ±3.6	97.9 ±5.2*	98.9 ±2.2*	96.9 ±6.1*

[a] Values are mean percent recovery relative to day 0 with standard deviations. An asterisk (*) indicates significant difference from day 0 mean ($P < 0.05$).

Freezer Storage Stability of Herbicides in Stream Water Samples

Mean percent recoveries for each herbicide from the Weogufka Creek water sample after 3, 6, and 12 months of freezer storage are given in table 5. As with the ambient temperature results, the freezer storage recoveries are excellent for both water samples in spite of the numerous significant mean differences. Means across all herbicides confirm that freezer storage is acceptable for up to one year.

Freezers must be monitored around the clock to prevent accidental thawing due to mechanical failure or extended power outages. Losses due to bottle cracking can be prevented by leaving at least 10% unfilled volume for expansion and by not stacking bottles tightly together.

Tubing-Related Carryover of Herbicides in Weogufka Creek Water Samples

Carryovers detected for each herbicide using the Weogufka Creek water sample are given in table 6. Each herbicide showed about 1% carryover in the first blank collected after spikes. No carryover was detected in the second or third blanks for any of the tested herbicides in either stream water matrix. If adsorption onto the inner tubing walls were occurring, then hydrophobic herbicides like sulfometuron methyl and triclopyr BE ester should exhibit greater wall adhesion and carryover than more water-soluble herbicides like imazapyr and hexazinone. Tubing wall contact times during a normal auto-sampler draw and back-flush cycle are brief. A 9.1 m length of 0.95 cm i.d. PVC suction tubing plus 104 cm of 0.95 cm i.d. silicone pump tubing has an internal volume of 726 mL. At a standard peristaltic pump draw rate of 3.5 L min^{-1} the contact time of any given 1 mL volume of water is

Table 5. Percent herbicide recoveries from stream water samples following freezer storage at < –15°C (mean values; n = 4).[a]

	Weogufka Creek Water			Escambia Stream Water		
Herbicide	Month 3	Month 6	Month 12	Month 3	Month 6	Month 12
Imazapyr	97.8 ±0.7*	102.4 ±0.8*	94.9 ±1.8*	93.9 ±0.5*	95.8 ±0.8*	93.9 ±0.2*
Hexazinone	97.1 ±0.4*	95.2 ±2.5*	94.4 ±0.7*	100.4 ±2.0	97.0 ±3.8	93.4 ±3.7*
Metsulfuron methyl	108.8 ±0.3*	131.0 ±0.8*	95.5 ±1.2*	99.2 ±0.2	99.5 ±0.6	97.0 ±0.4*
Sulfometuron methyl	105.9 ±0.5*	99.2 ±0.4*	103.4 ±0.1*	101.1 ±2.5	91.6 ±1.4*	84.5 ±2.8*
Triclopyr	95.9 ±0.4*	93.6 ±0.2*	95.7 ±0.5*	99.0 ±0.5	99.8 ±0.4	101.9 ±0.2*
Triclopyr BE ester	98.3 ±1.0	101.9 ±1.4	105.8 ±2.0*	99.9 ±1.2	99.8 ±0.9	104.4 ±1.6*
Mean (all herbicides)	100.6 ±5.0	103.9 ±12.9*	98.3 ±4.8*	98.9 ±2.7	97.2 ±3.4*	95.8 ±6.8*

[a] Values are mean percent recovery relative to day 0 with standard deviations. An asterisk indicates significant difference from day 0 mean ($P < 0.05$).

Table 6. Percent recovery of herbicides in automatic sampler blanks following spikes (mean values; $n = 4$).[a]

	Weogufka Creek Water			Escambia Stream Water		
Herbicide	First Blank	Second Blank	Third Blank	First Blank	Second Blank	Third Blank
Triclopyr	1.1 ±0.1	ND	ND	0.8 ±0.2	ND	ND
Triclopyr ester	ND	ND	ND	ND	ND	ND
Sulfometuron methyl	0.8 ±0.2	ND	ND	1.1 ±0.2	ND	ND
Metsulfuron methyl	0.8 ±0.1	ND	ND	1.2 ±0.2	ND	ND
Imazapyr	ND	ND	ND	0.9 ±0.1	ND	ND
Hexazinone	0.8 ±0.3	ND	ND	1.3 ±0.2	ND	ND

[a] Values are mean percent recovery relative to mean spike recovery with standard deviations. ND = not detected above MDL.

only 13 s. This suggests that physical retention of water droplets in the tubing rather than surface leaching from the tubing was responsible for the slight carryover found in the first blanks.

The intent of this study was to replicate and test sampling protocols as we practice them under remote field conditions. Multiple back-flushes prior to sample collection were not used because of excessive battery drain. The abrupt falloffs in herbicide residue levels modeled above have never been observed in field studies. Actual residue levels have tapered off too gradually for such slight carryovers to be detectable.

SUMMARY AND CONCLUSIONS

This study was performed to evaluate the effects, if any, of temporary ambient and long-term freezer storage of stream water samples in HDPE sample bottles and PVC automatic sampler tubing used in environmental fate studies of forestry herbicides. This work studied the pre-analysis collection and storage of stream water samples from forest watersheds during environmental fate studies. Recoveries of six forestry herbicides from two stream waters were monitored before and after sample withdrawal, ambient temperature standing, and freezer storage. None of the six herbicides exhibited recovery loss from either water matrix after six days at ambient temperature or one year of freezer storage. None of the six adsorbed on auto-sampler suction tubing or peristaltic pump tubing during typical sample withdrawal and back-flush cycles.

ACKNOWLEDGEMENTS

The authors wish to acknowledge Ronald Tucker, Supervisory Forestry Technician, for his expert assistance in site selection and collection of the stream water samples from the Escambia Experimental Forest.

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