Invasive Plant Science and Management 2017 10:277–283

Sequential Aminopyralid and Imazapyr Applications for Japanese Knotweed (*Fallopia japonica*) Management

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Japanese knotweed is an invasive plant that occurs along waterways, highways, abandoned agricultural land, and other disturbed areas. It reduces plant diversity and can increase shoreline erosion. An experiment was conducted in Bible Hill and Antigonish, NS, Canada to evaluate early POST aminopyralid at 120 g ae ha^{-1} and POST imazapyr applications at 720 g ae ha^{-1} at maximum shoot height, flowering, senescence, maximum height + flowering, maximum height + senescence, flowering + senescence, and maximum height + flowering + senescence. Early POST aminopyralid only provided 10% to 15% control at 52 wk after treatment (WAT) whereas 83% to 100% control occurred following imazapyr applications at all application timings. Percent control at 2, 4, and 8 WAT tended to be higher where imazapyr followed aminopyralid. By 52 WAT, equivalent damage ratings and reductions in stem density occurred at both sites in all plots where imazapyr was applied. The use of aminopyralid or multiple imazapyr applications provided no additional benefit over a single imazapyr application. We conclude that early POST aminopyralid suppresses knotweed growth, which should facilitate late-season imazapyr applications, especially in large stands.

Nomenclature: Aminopyralid; imazapyr; Japanese knotweed, *Polygonum cuspidatum* Sieb. & Zucc., syn. *Fallopia japonica* (Houtt.) Dcne. POLCU.

Key words: Invasive species, early POST herbicide, riparian zone.

Japanese knotweed (*Polygonum cuspidatum*) is a resilient, invasive nonnative plant species in North America. It was originally introduced as an ornamental plant (Beerling et al. 1994) and has become an aggressive invader of natural areas, where it can outcompete with and eliminate other plant species (Aguilera et al. 2010; Gerber et al. 2008; Murrell et al. 2011). Areas characterized by moderate disturbance and ample resource availability, such as roadsides and waterways, are prone to invasion by Japanese knotweed (Beerling et al. 1994; Claeson and Bisson 2013; Dommanget et al. 2013). It reproduces predominantly by clonal propagation through extension of rhizomes (Beerling et al. 1994; Smith et al. 2007) that can grow downward to a depth of 2 m and extend several meters outward from existing stands before producing new shoots (Child and Wade 2000). Shoots generally emerge early in the season (Beerling 1990), with emergence thought to be regulated by temperature due to variations in emergence timing across locations and years (Beerling et al. 1994; Dauer and Jongejans 2013). New shoots emerge in Nova Scotia, Canada in early spring (April to May), produce flowers in August, and are killed by frost in October (Larsen 2013).

Management of Japanese knotweed generally relies on a combination of methods (Bram 2002), though effectiveness of different management approaches is variable (Delbart et al. 2012). Mechanical or physical controls include using tarps or wire mesh to stunt the growth of young shoots and cutting stems at different growth stages to inhibit allocation of resources to the rhizomes (Colleran and Goodall 2014; Delbart et al. 2012; Dommanget et al. 2013; Larsen 2013). These methods, however, can be expensive and labor-intensive, require extensive follow-up, and may even promote spread of the plant (Beerling 1990). Biological control with insect and disease organisms is promising (Grevstad et al. 2013; Shaw et al. 2009), though this approach has not been widely implemented. Symplastic herbicides are generally the most efficient and effective means of managing Japanese knotweed due to translocation to rhizomes following foliar applications (Bashtanova et al. 2009), with glyphosate, imazapyr, and several synthetic auxins (aminopyralid, 2,4-D, dicamba, and triclopyr) most

DOI: 10.1017/inp.2017.31

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Management Implications

Japanese knotweed is a common invasive plant species that occurs along waterways, in parks, in abandoned agricultural fields, and in other disturbed areas. An established stand of knotweed outcompetes other vegetation and reduces localized species diversity. It also destabilizes the banks of waterways and can make them more susceptible to erosion. A variety of management techniques have been evaluated, but herbicides are consistently the most cost-effective approach. Our results indicate that an application of aminopyralid at 120 g ae ha^{-1} when shoots are approximately 30-cm tall will not adequately control knotweed but will suppress shoot growth. POST imazapyr applications at 720 g ae ha^{-1} are effective when applied any time between maximum height and just before shoot senescence in the fall. By 52 WAT, all imazapyr application timings provided similar levels of control, with multiple imazapyr applications providing no additional benefit. In large stands, early POST aminopyralid applications may suppress knotweed growth and facilitate late-season imazapyr applications. This technique is not recommended along waterways, as it kills most vegetation, leaving areas of bare soil that would be susceptible to erosion.

commonly evaluated (Bashtanova et al. 2009; Delbart et al. 2012; Hagen and Dunwiddie 2008; Johnson et al. 2010). Foliar applications at optimum timings for herbicide translocation to rhizomes are difficult, however, due to the height of established knotweed plants (Weston et al. 2005). Assimilate translocation to rhizomes is greatest in late summer and early fall (August to September) (Price et al. 2002), when established knotweed stems often exceed 3 m in height (Larsen 2013). Cutting of stems in early summer followed by treatment of regrowth can improve ease of herbicide applications (Johnson et al. 2010) but is labor-intensive and requires careful disposal of clipped stems due to risk of plant dispersal from stem fragments (De Waal 2001), particularly in riparian habitats (Brock and Wade 1992). Alternatively, initial growth of knotweed stems can potentially be suppressed with early-season herbicide applications. In previous work, Kay (2003) indicated potential suppressive effects of early POST applications of the diglycolamine salt of the synthetic auxin herbicide dicamba on knotweed, which reduced shoot density and, to some extent, shoot height approximately 1 mo after application. Further work with synthetic auxin herbicides as early-season treatments on knotweed are recommended (Clements et al. 2016), but results from field experiments to date are limited.

Synthetic auxins are generally less effective than other herbicides on Japanese knotweed (Delbart et al. 2012) but may improve management when used in conjunction with other herbicides, as they cause less damage to some non-target plants in comparison to other active ingredients (Johnson et al. 2010; Rudenko and Hulting 2010). In particular, aminopyralid has gained increasing use in Nova Scotia due to recent success with this product for management of wild chervil [*Anthriscus sylvestris* (L.) Hoffm.] on dykelands (Beaton 2014) and lack of availability of alternative products with similar efficacy (R Hoeg, Nova Scotia Department of Agriculture, personal communication). Aminopyralid is a synthetic auxin herbicide commonly used for early POST and POST control of annual and perennial broadleaf weeds in pastures, rangelands, and non-crop areas (Enloe et al. 2007, 2008; Ferrel et al. 2006). Weed control is generally most consistent following POST applications (Kyser et al. 2011), but early POST applications control perennial weed seedlings (Ferrel et al. 2006) and may suppress developing shoots of some perennial plants (Seefeldt et al. 2013). Aminopyralid efficacy on Japanese knotweed is generally more consistent than that observed with other synthetic auxin herbicides (Delbart et al. 2012; Rudenko and Hulting 2010), but control is not as good as observed with products such as imazapyr and glyphosate. It is possible, however, that early season aminopyralid applications to young, rapidly growing knotweed stems may suppress stem growth and facilitate later herbicide applications at phenological stages conducive to assimilate translocation to rhizomes. The objectives of this research were to determine (1) the effect of early POST aminopyralid applications on Japanese knotweed shoot growth and (2) whether early POST aminopyralid applications improve control of Japanese knotweed when used in conjunction with POST applications of imazapyr at multiple growth stages.

Materials and Methods

Species Description. The plants at both sites were identified as Japanese knotweed, and not the hybrid Bohemian knotweed (*Polygonum × bohemicum* (J. Chrtek & Chrtková) Zika & Jacobson [*cuspidatum × sachalinense*]). This was established using several identification keys (Barney et al. 2006; Child and Wade 2000; Wilson 2007; Zika and Jacobson 2003) to differentiate various morphological characteristics such as canopy height (2- to 3-m tall), leaf shape (cuspidate apex and flattened base), leaf size (less than 15-cm long and 12-cm wide), leaf hairs (glabrous), and flowers (female only) (Larsen 2013). Leaf samples were sent to Agriculture and Agri-Food Canada in Lethbridge, AB, in September 2010 for analysis using amplified fragment length polymorphism (AFLP-PCR). They concluded that the specimens were Japanese knotweed *Polygonum cuspidatum* (R Bourchier, personal communication).

Site Description. Experiments were conducted at Antigonish and Bible Hill, NS, in 2011 and 2012. The Antigonish site was located along the south side of the Rights River (45.622°N, 61.972°W), which flows through the town of Antigonish, NS, Canada. The soil at the site is a

https://doi.org/10.1017/inp.2017.31 Published online by Cambridge University Press

Table 1. Herbicide treatments for Japanese knotweed at two sites in Nova Scotia, Canada in 2011.

		Application date			
Herbicide	Site	Emergence	Maximum growth	Flowering	Presenescence
Aminopyralid (Milestone™)	Antigonish	May 18	—	—	—
	Bible Hill	May 12	—	—	—
Imazapyr (Arsenal®)	Antigonish		July 20	August 30	October 12
	Bible Hill		July 11	August 23	October 12

silt loam, and research plots were situated at approximately 1 to 2 m above sea level in Japanese knotweed patches found along both sides of the river. A soil analysis of particle-size distribution, as described by Sheldrick and Wang (1993) and Brewster (2001), measured 62% sand, 13% clay, and 25% silt. The Bible Hill site was located along the Salmon River, which separates the towns of Bible Hill and Truro, NS (45.371°N, 63.277°W). The soil at the site is a sandy loam, and research plots were situated approximately 13 m above sea level in Japanese knotweed patches along one side of the river. Part of the site had been used as pastureland but had been left fallow for several years before our study was initiated. A soil analysis of particle-size distribution, measured 70% sand, 15% clay, and 15% silt.

Experimental Design. The experiment was a randomized complete block design with three blocks and a two by eight factorial treatment arrangement. The factors were early POST aminopyralid (Milestone™, Dow AgroSciences, Indianapolis, IN 46268) applications (presence vs. absence) and POST imazapyr (Arsenal®, BASF, Research Triangle Park, NC 27709) applications (none, maximum stem height [M], flowering [F], presenescence [S], M + F, M + S, F + S, and M + F + S). Plots were 2 by 2 m with a 2-m buffer between each plot. The buffers between plots were mowed throughout the season to permit travel throughout the site. Aminopyralid was applied at 120 g ae ha^{-1} using a CO_2-pressurized research plot sprayer (Bellspray, Opelousas, LA 70570) outfitted with four 8002VS nozzles (TeeJet®, Springfield, IL 62703) and calibrated to deliver a water volume of 200 L ha^{-1} at 270 kPa. Aminopyralid applications were made on May 12, 2011, at Bible Hill and May 18, 2011, at Antigonish, when emerged stems were approximately 30-cm tall. Imazapyr was applied at a rate of 720 g ae ha^{-1} in 1,200 L ha^{-1} of water in all plots with 0.5% v/v nonionic surfactant (Agral® 90, Syngenta Canada, Guelph, ON N1G 4Z3, Canada). A large spray volume was used to ensure coverage of the thick canopy. Imazapyr was applied using a 15-L Solo® backpack sprayer (Solo®, Newport News, VA 23605) equipped with a single TeeJet® 11004VP nozzle, and all sides and the top of each 4-m^2 knotweed plot were treated. The same spray volume was applied to all plots regardless of canopy size. Application timings for imazapyr treatments are provided in Table 1.

Data Collection. Visual estimates of herbicide injury were conducted using a 0 to 100 scale where 0 represents no plant damage and 100 represents complete shoot death. Ratings were conducted at 2, 4, 8, and 52 wk after the final POST herbicide application treatment (WAT). Stem density was counted on a whole-plot basis at 2, 8, and 52 WAT. Stem height was determined on 5 randomly chosen knotweed stems in each plot at 8 and 52 WAT. Knotweed aboveground biomass was measured at 52 WAT by cutting all knotweed stems in each plot and weighing them to the nearest gram on a portable scale in the field to determine fresh weight.

Data Analysis. Data were analyzed using PROC MIXED in SAS v. 9.3 (SAS Institute, Cary, NC 27513) with block as a random factor. Sites were analyzed separately. Normality and constant variance assumptions were verified. In some cases, transformations were used to achieve normality, and back-transformed means are presented. For significant effects, the least-squares means were compared with Tukey's adjustment ($P \leq 0.05$). The subroutine pdmix800.sas (Saxton 1998) was used to provide letter groups. The 2011 data were collected at multiple time points and were analyzed using repeated measures in SAS with week as the time factor. The data for 2012 were collected on a single date, and the analysis was similar, except it was not analyzed as a repeated measure.

Results and Discussion

At both sites, early POST aminopyralid and POST imazapyr increased knotweed damage ratings, and the effect changed over time (Table 2). Aminopyralid on its own did not adequately control knotweed, with only 10% to 15% suppression at 52 WAT (Table 3). However, percent control at all POST imazapyr application timings at 2, 4, and 8 WAT tended to be higher at both sites when the application followed early POST aminopyralid, although the differences

https://doi.org/10.1017/inp.2017.31 Published online by Cambridge University Press

Table 2. Significance levels (P-values) for all combinations of herbicide treatments on damage assessments at Antigonish and Bible Hill, NS, in 2011.

Site	Effect[a]	Damage rating	Density	Height	Biomass
Antigonish					
	Week	<0.0001	0.0001	<0.0001	—
	Early POST	<0.0001	0.0001	<0.0001	0.9907
	Week*early POST	<0.0001	0.0120	0.0378	—
	POST	<0.0001	<0.0001	0.0024	0.0199
	Week*POST	<0.0001	0.0002	<0.0001	—
	Early POST*POST	<0.0001	0.0019	0.2735	0.0839
	Week*early POST*POST	<0.0001	0.0017	0.7897	—
Bible Hill					
	Week	<0.0001	<0.0001	<0.0001	—
	Early POST	<0.0001	0.0016	<0.0001	0.0563
	Week*early POST	<0.0001	0.0976	0.0667	—
	POST	<0.0001	<0.0001	0.0022	<0.0001
	Week*POST	<0.0001	<0.0001	<0.0001	—
	Early POST*POST	0.0008	0.7675	0.0011	0.3950
	Week*early POST*POST	0.0654	0.2941	0.9825	—

[a] Effects were: week (2, 4, and 8 wk after treatment), early POST (aminopyralid when stems were 30-cm tall), and POST (imazapyr at multiple growth stages), as well as the interaction.

were not always significant. We also found a reduction in stem density at 52 WAT in Antigonish (Table 4) and 2 WAT in Bible Hill (Table 5) where early POST aminopyralid was applied alone compared with the nontreated control as well as a reduction in height at both sites (unpublished data). We cannot fully explain the increased control observed when imazapyr followed aminopyralid, but given that aminopyralid applications tended to reduce stem density and stem height of Japanese knotweed, it is possible that aminopyralid applications facilitated increased shoot coverage by imazapyr. Adequate coverage is especially difficult to achieve in large infestations, and in this type of situation an early aminopyralid application might play a critical role in facilitating POST herbicide applications.

Table 3. The effect of early POST aminopyralid applications and POST imazapyr applications at maximum knotweed height, flowering, maximum height + flowering, presenescence, maximum height + presenescence, flowering + presenescence, and maximum height + flowering + presenescence on percent Japanese knotweed control at 2, 4, 8 and 52 wk after the final POST herbicide application (WAT) at Antigonish and Bible Hill, NS, in 2011.

	POST	Antigonish[a]				Bible Hill[a]			
Early POST	Timing	2 WAT	4 WAT	8 WAT	52 WAT	2 WAT	4 WAT	8 WAT	52 WAT
		% control							
No aminopyralid	Nontreated[b]	0	0	0	0	0	0	0	0
	Maximum height (M)	10 o	16 mno	33 ijk	90 abc	10 k	10 k	20 ijk	87 abc
	Flowering (F)	13 no	20 lmn	60 d-g	93 ab	10 k	20 ijk	43 gh	86 a-d
	M + F	30 jkl	36 h-k	87 a-d	100 a	20 ijk	30 hij	53 e-h	97 a
	Senescence (S)	23 klm	—	—	83 a-e	13 jk	—	—	87 abc
	M + S	83 a-e	—	—	97 a	43 gh	—	—	100 a
	F + S	60 d-g	—	—	97 a	43 gh	—	—	100 a
	M + F + S	90 abc	—	—	100 a	56 efg	—	—	100 a
Aminopyralid	Nontreated	56 efg	49 ghi	60 d-g	10 o	50 fgh	47 fgh	43 gh	15 jk
	M	53 fgh	60 d-g	60 d-g	97 a	53 e-h	57 d-g	57 d-g	190 ab
	F	46 g-j	49 ghi	62 c-g	90 abc	50 egh	47 fgh	73 a-f	90 ab
	M + F	76 a-f	79 a-f	97 a	93 ab	57 d-g	62 b-g	80 a-e	90 ab
	S	33 ijk	—	—	90 a-d	40 ghi	—	—	86 abc
	M + S	90 abc	—	—	93 ab	90 ab	—	—	97 a
	F + S	63 b-g	—	—	93 ab	60 c-g	—	—	100 a
	M + F + S	97 a	—	—	100 a	87 abc	—	—	100 a

[a] Mean values within a site with the same letter were not significantly different using Tukey's test at $P \leq 0.05$.
[b] The nontreated control was removed from the analysis.

Table 4. The effect of early POST aminopyralid applications and POST imazapyr applications at maximum knotweed height, flowering, maximum height + flowering, presenescence, maximum height + presenescence, flowering + presenescence, and maximum height + flowering + presenescence on Japanese knotweed density at 2 and 52 wk after the final POST herbicide application (WAT) at Antigonish, NS, in 2011.

	POST	Shoot density[a]	
Early POST	Timing	2 WAT	52 WAT
		stems m^{-2}	
No aminopyralid	Nontreated	19 ab	33 a
	Max. height (M)	16 bcd	4 c-f
	Flowering (F)	17 bc	2 ef
	M + F	18 b	0 f
	Senescence (S)	12 b-f	3 c-f
	M + S	13 b-f	0 f
	F + S	12 b-f	1 f
	M + F + S	12 b-f	0 f
Aminopyralid	Nontreated	16 b-e	12 b-f
	M	14 b-f	1 ef
	F	14 b-f	3 c-f
	M + F	6 b-f	2 def
	S	14 b-f	11 b-f
	M + S	2 ef	1 ef
	F + S	7 b-f	3 def
	M + F + S	1 f	0 f

[a] Mean values with the same letter were not significantly different using Tukey's test at $P \leq 0.05$.

Table 5. The effect of early POST aminopyralid applications and POST imazapyr applications at maximum knotweed height, flowering, maximum height + flowering, presenescence, maximum height + presenescence, flowering + presenescence, and maximum height + flowering + presenescence on Japanese knotweed density at 2 and 52 wk after the final POST herbicide application (WAT) at Bible Hill, NS, in 2011.

	Shoot density[a]	
Herbicide	2 WAT	52 WAT
	stems m^{-2}	
No aminopyralid	15 a	5 a
Aminopyralid	9 b	4 a
Nontreated	14 ab	22 a
Max. height (M)	—	3 c-f
Flowering (F)	11 bc	2 def
M + F	10 bcd	1 ef
Senescence (S)	9 b-e	4 c-f
M + S	6 b-f	1 ef
F + S	6 b-f	0 ef
M + F + S	7 b-f	0 f

[a] Mean values with the same letter were not significantly different using Tukey's test at $P \leq 0.05$.

Percent control at 2, 4, and 8 WAT where no aminopyralid was applied tended to be higher when imazapyr was applied at maximum height + flowering growth stages versus a single application at maximum height (Table 3). This difference did not occur where aminopyralid was applied. Damage tended to increase from 2 to 4 WAT in all treatments. At 52 WAT, all treatments had 83% to 100% control at both sites. These results suggest that POST imazapyr is as effective as early POST aminopyralid followed by POST imazapyr 1 yr after treatment. All application timings for imazapyr 1 yr after application resulted in similar damage levels, and sequential applications provided no added benefit. These results are somewhat surprising, as Price et al. (2002) determined that freshly synthesized carbohydrates predominantly remain in the stem in spring and early summer (May and June), with sharp increases in translocation of carbohydrates to the rhizome in late summer or presenescence. Similarly, Bashtanova et al. (2009) argue that products such as glyphosate are not effective because of limited translocation to the rhizome until the fall, when there is limited protein synthesis. In contrast, Seiger and Merchant (1997) reported season-long allocation to belowground biomass in Japanese knotweed. Imazapyr was equally effective at all application timings 1 yr after application in our research, suggesting that it was translocated to the rhizomes at all growth stages. Our results are in agreement with Seiger and Merchant (1997) and indicate that opportunities for belowground herbicide translocation in Japanese knotweed may exist earlier in the season than previously thought.

Knotweed density varied with time and was affected by both herbicide types at both sites (Table 2). There was a significant early POST by POST interaction at Antigonish but not at Bible Hill. Density did not change over a 1-yr period in the nontreated controls at either site, though density tended to increase at Antigonish (Tables 4 and 5). In Antigonish, where aminopyralid was not applied, there were no differences in density 2 wk after the final POST application (Table 4). Where aminopyralid was applied, densities were lower than the nontreated control, where imazapyr was applied at maturity + senescence and maturity + flowering + senescence. All POST herbicide treatments reduced knotweed density at 52 WAT compared with the nontreated control at both sites.

Shoot heights in the plots that received aminopyralid were 21% to 42% shorter than those that did not (unpublished data) at 2, 4, and 8 WAT. POST imazapyr applications did not affect height at 2 WAT, but heights at both sites by 52 WAT were significantly stunted by all

Table 6. The effect of POST imazapyr applications at maximum knotweed height, flowering, maximum height + flowering, presenescence, maximum height + presenescence, flowering + presenescence, and maximum height + flowering + presenescence on Japanese knotweed height at 2 and 52 wk after the final POST herbicide application (WAT) and knotweed fresh shoot biomass at Bible Hill and Antigonish, NS, in 2011.

Herbicide	Antigonish[a]		Bible Hill[a]		Antigonish[a]	Bible Hill[a]
	2 WAT	52 WAT	2 WAT	52 WAT	52 WAT	52 WAT
	cm				kg ha^{-1}	
Nontreated	96 abc	166 a	127 ab	159 a	16 a	19 a
Max. height (M)	140 a	34 c	149 a	64 c	0 b	2 b
Flowering (F)	149 a	42 c	140 ab	73 bc	0 b	2 b
M + F	133 a	27 c	153 a	60 c	1 ab	1 b
Senescence (S)	148 a	94 abc	160 a	64 c	7 ab	2 b
M + S	131 a	39 c	159 a	27 c	1 ab	1 b
F + S	150 a	44 bc	155 a	29 c	1 ab	1 b
M + F + S	131 a	—	152 a	23 c	15 ab	1 b

[a] Mean values with the same letter were not significantly different using Tukey's test at $P \leq 0.05$.

herbicide application timings (Table 6). All POST imazapyr application timings caused a significant reduction in biomass at 52 WAT (Table 6).

The results indicate that early POST aminopyralid applications did not increase the level of control achieved by 52 WAT. However, aminopyralid applications suppressed knotweed height. Imazapyr is very effective over a wide range of growth stages. Early percent control was greatest with applications at maximum height + flowering < maximum height + senescence = maximum height + flowering + senescence. This difference disappeared by 52 WAT. We conclude that POST imazapyr is an effective management for knotweed across a range of application timings, and early POST aminopyralid may suppress knotweed growth and facilitate POST applications.

Acknowledgments

Funding was provided by the Nova Scotia Department of Transportation and Infrastructure Renewal.

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Received December 12, 2016, and approved April 4, 2017.

Associate Editor for this paper: Steven S. Seefeldt, University of Alaska, Fairbanks.