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Author for correspondence:
Patrick J. Minogue, Associate Professor, University of Florida, North Florida Research and Education Center, 155 Research Road, Quincy, FL 32351. Email: pminogue@ufl.edu

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Comparison of aminocyclopyrachlor to standard herbicides for basal stem treatment of *Eucalyptus benthamii*

Patrick J. Minogue[1] and Kimberly A. Lorentz[2]

[1]Associate Professor, University of Florida, North Florida Research and Education Center, Quincy, FL, USA and [2]Graduate student, University of Florida, School of Forest Resources and Conservation, Gainesville, FL, USA

Abstract

Eucalyptus species are grown for fiber, fuel, and other uses on more than 17.8 million ha worldwide, yet some species are considered invasive and may have adverse environmental or social impacts outside their native range. Aminocyclopyrachlor (AMCP) and standard applications of imazapyr and triclopyr herbicides were compared for eucalyptus control using a basal stem application method. At 6 and 12 mo after treatment (MAT), basal stem applications using 5% (vol/vol) AMCP (120 g ae L^{-1}) in methylated soybean oil (MSO) resulted in 97% to 99% eucalyptus crown reduction and generally provided greater control across all diameter classes than standard treatments of 28% imazapyr (240 g ae L^{-1}) or 75% triclopyr ester (480 g ae L^{-1}). AMCP at 5% was as effective as 40% vol/vol. Increases in stem live height at 24 MAT suggest that the effect of triclopyr ester basal stem treatment may be impermanent. AMCP treated trees did not have regrowth by 24 MAT.

Introduction

Members of the diverse *Eucalyptus* genus (family Myrtaceae) are native to Australia and the bordering islands of Polynesia (Gonzáles-Orozco et al. 2014) and more than 70 species are naturalized elsewhere (Rejmánek and Richardson 2011). Because of their adaptability and fast growth rates, eucalyptus species and hybrids have been grown for fiber, fuel, landscaping mulch, essential oils, phytoremediation purposes, and as ornamental plants (Davidson 1993; Rockwood 2012) on more than 17.8 million ha worldwide (Pires et al. 2013). In the southeastern United States, eucalyptus has historically been used for pulp, mulch, and windbreaks. Now, there is renewed interest in planting cold-tolerant species, hybrids, and genetically modified stock to supply potential bioenergy markets. *Eucalyptus benthamii* is among the more promising eucalyptus species for wide-scale planting in the southeastern United States because of its cold hardiness, success in plantations under a variety of conditions, and fast growth rates (Zalesny et al. 2011).

Concerns have been raised over the potential for eucalyptus to become invasive (Callaham et al. 2013; Gordon et al. 2011; Lorentz and Minogue 2015a). Eight eucalyptus species are considered invasive in various regions in the world (Rejmánek and Richardson 2013), but *E. benthamii* is not among them. However, the ability of this species to withstand cold weather could contribute to its potential invasiveness at a geographic scale. Where eucalyptus species have invaded, they have had undesirable economic and ecological impacts, including greater fire intensity (Pagni 1993); reduction of natural river flows (Le Maitre et al. 2002); alteration of the native faunal composition and density (Sax 2002); and negative changes in plant richness, diversity, and structural attributes (Tererai et al. 2013). Contributing to the extensive propagation of eucalyptus is their perennial growth form, prolific seed production, low incidence of disease, insect resistance, drought tolerance, rapid growth rate, and adaptability to infertile soil (Booth 2013). As eucalyptus are planted over greater areas to meet a growing fiber and energy wood demand (Dougherty and Wright 2012; Hodges et al. 2010), the risk of invasion will also be driven by potentially high production of seed propagules, dispersed stands, and plantings adjacent to disturbed or natural areas (Lorentz and Minogue, 2015b).

The Invasive Species Advisory Committee, a group of non-federal experts and stakeholders established to provide advice on invasive species issues to the interagency National Invasive Species Council (Federal Register 1999), made nine recommendations for federal biofuels programs to minimize the risk of bioenergy crop escape into the surrounding environment. Their recommendations include the need to establish protocols for rapid removal of bioenergy crops should they disperse into surrounding areas or become abandoned and unwanted populations (NISC 2009). Herbicides are a relatively effective and inexpensive tool that can be used to manage eucalyptus seedlings occurring from natural recruitment or for the removal of abandoned stands of mature trees. To prepare for potential management of invasive eucalyptus in the southeastern United States, effective chemical control methods need to be established.

Existing recommendations for chemical control of naturalized eucalyptus are not refined. They generally entail cut stump, basal frill (also known as cut stem treatment or hack and squirt), or basal stem (also known as basal bark) applications using concentrated herbicide solutions or emulsions containing the active ingredients triclopyr, imazapyr, or glyphosate. Individual plant treatments using these broad-spectrum herbicides are preferred for their ease of use and targeted application. Triclopyr and glyphosate may be applied to the stems of target vegetation with minimal impact to nearby vegetation, because these herbicides are not readily root-absorbed from the soil (Senseman 2007). However, imazapyr is a soil-active herbicide, thus injury to nontarget vegetation is more likely (Little and Shaner 1991).

Cut stump applications of these herbicides are also widely used in eucalyptus plantations for silviculture objectives such as managing tree disease, replacing rootstocks with improved cultivars, or improving stand vigor between 5- to 10-year harvesting cycles. Following felling, most species planted in commercial plantations regenerate from vigorous stump spouts. Cold-hardy species such as *Eucalyptus amplifolia* Maiden et Cambage and *Eucalyptus macarthurii* H. Deane & Maiden, widely planted in the southeastern United States, southern Brazil, and South Africa (Minogue et al. 2018), are proving difficult to kill by existing cut stump methods (Little and van den Berg 2007).

Current herbicide recommendations for eucalyptus control include triclopyr, glyphosate, and imazapyr, and these are effective in cut-stump and cut-stem applications (Bossard et al. 2000; Moore 2008). However, complete control of eucalyptus is rarely achieved from a single herbicide treatment and reapplication to control sprouting is often necessary (Bachelard et al. 1965; Bossard et al. 2000; Little 2003; Little and van den Berg 2006; Morze 1971), potentially taking up to three herbicide treatments to provide control (Bossard et al. 2000). Furthermore, variable tolerance across species and varying effectiveness over application timings for commonly used herbicides make it difficult to offer recommendations on its use (Bachelard et al. 1965; Morze 1971). Few guidelines consider tree size and vigor, which are also important for the success of herbicide treatments (Morze 1971).

Aminocyclopyrachlor (AMCP) was developed by DuPont for use in non-crop areas such as rights-of-way, turf, and range and natural areas (Anomymous 2009). A pyrimidine carboxylic acid herbicide, it is structurally similar to pyridine carboxylic acid herbicides such as aminopyralid, picloram, and triclopyr. Its synthetic auxin mode of action interferes with normal plant growth (USDA 2012). AMCP is effective at low application rates for control perennial woody plants including kudzu [*Pueraria montana* (Lour.) Merr.; Minogue et al. 2011], largeleaf lantana (*Lantana camara* L.; Ferrell et al. 2012) and Chinese tallow [*Triadica sebifera* L. (Small); Enloe et al. 2015].

In an early screening for forest vegetation management Yeiser et al. (2011) compared the use of 2.5%, 5%, 10%, and 15% AMCP (120 g ae L^{-1}) to 30% triclopyr ester (480 g ae L^{-1}) applied to the cut stems of Chinese tallow [*Triadica sebifera* L. (Small)], sweetgum (*Liquidambar styraciflua* L.), and yaupon (*Ilex vomitoria* Aiton). At 18 mo after treatment (MAT) 10% AMCP and triclopyr resulted in 100% control of Chinese tallow. Triclopyr controlled sweetgum the best and gave 100% control of yaupon. These results showing differences in species susceptibility warrant further testing of AMCP rate response for potential silvicultural or invasive eucalyptus management options. The objective of this research was to compare the efficacy of four AMCP rates to two standard herbicides for control of *E. benthamii* using basal stem applications.

Materials and Methods

Study Sites

In September 2011, two identical studies were initiated in separate 0.2-ha *E. benthamii* plantations at the University of Florida, North Florida Research and Education Center, south of Quincy (30.55°N, 84.60°W). This location has a temperate climate with highest temperatures in July (mean 27 C), lowest temperatures in January (mean 10 C), and 143 cm mean annual precipitation (NOAA 2002). The prevalent soil series at both locations is Orangeburg fine sandy loam (fine-loamy, kaolinitic, thermic Typic Kandiudults; USDA-NRCS 2020), but soils at the north study site were highly eroded, to the extent that the typic sandy surface horizon was absent. Hereafter, the two study sites are referred to as eroded and non-eroded. In June 2009, 16-wk-old containerized seedlings of *E. benthamii* were hand planted in rows (2.4 m between rows) at 1.5-m intervals in the row. In a 2009 study in these plantations (Minogue et al. 2018), trees received different levels of weed control during the establishment year. This resulted in a range of tree sizes, from less than 1 to 10 m in height and from 4 to 20 cm in basal stem diameter (BSD) at groundline, when measured prior to application of herbicide treatments in late October 2011. Trees were generally smaller on the eroded site.

Basal Stem Treatments

The experimental design was a randomized complete block with seven treatments (Table 1). At each study site 140 healthy trees were used as experimental units, leaving at least one buffer tree between them. Because tree size could influence treatment comparisons, treatment trees were ranked according to basal diameter and divided sequentially into 20 groups of seven trees (blocks). Each block represented the range of tree diameter, with the tallest tree in block one, second tallest in block two, and so on. Basal stem applications of AMCP (120 g ae L^{-1}) in the form of DPX MAT28-159 OL, an oil-based liquid formulation developed for use in basal stem application (Anonymous 2009), were tested at four concentrations (5%, 10%, 20%, and 40% vol/vol formulated product). Because AMCP was an experimental herbicide at the time of this study, the range of treatment concentrations was selected based on evidence from testing it on other woody plants (Edwards and Beck 2011; Wilson et al. 2011; Yeiser et al. 2011; J. Ferrell, University of Florida and M. Link, DuPont, personal communications, May 2011).

Comparison treatments included the standard basal stem applications 28% (vol/vol) imazapyr (240 g ae L^{-1}; Anonymous 2012) and 75% (vol/vol) triclopyr ester (480 g ae L^{-1}; Anonymous 2019) and a herbicide-free check (seed oil carrier only). All herbicides were thoroughly mixed with 100% methylated soybean oil, alkylphenol ethoxylate, as the carrier (Anonymous 2015). Five milliliters of herbicide/oil mixture per 2.5 cm groundline BSD were applied to the base of trees from 30 cm height to the groundline using a volumetric syringe. Preliminary testing indicated this volume was generally sufficient to wet the stem completely for the various diameter classes studied. Herbicide treatments were applied on November 4 and 5, 2011, and no rainfall occurred within 48 h following application.

Table 1. Basal stem treatments for *Eucalyptus benthamii* control using AMCP and standard herbicides in methylated seed oil carrier.[a,b]

Herbicide	Formulation	Herbicide concentration	Applied formulation concentration in oil	BSD specific dose
		g ae L^{-1}	% v/v	g ae per 2.5 cm BSD
AMCP	DPX MAT28-159	120	5	0.03
AMCP	DPX MAT28-159	120	10	0.06
AMCP	DPX MAT28-159	120	20	0.12
AMCP	DPX MAT28-159	120	40	0.24
Imazapyr	Stalker®	240	28	0.34
Triclopyr ester	Garlon® 4 Ultra	480	75	1.80
Nontreated check	MSO only	–	–	–

[a]Five milliliters of herbicide in oil mixture was applied per 2.5 cm BSD from 30-cm stem live height to groundline.
[b]Abbreviations: AMCP, aminocyclopyrachlor; BSD, basal stem diameter; MSO, methylated seed oil.

Tree Assessments

Each tree was assessed at 2, 6, and 12 MAT for percent crown reduction. As defined by Miller and Glover (1991), this Weed Science Society of America standard variable for forest herbicide research is a visual estimate of crown volume change relative to the pretreatment condition, and considers stem dieback, leaf necrosis, defoliation, and crown growth (growth indicated by negative crown reductions values). It is useful for making periodic assessments of crown volume changes in trees or shrubs during the period that the original pretreatment crown volume is evident. Estimates were made to the nearest 5 percent for values between 0% and 10%, and 90% and 100%; and to the nearest 10 percent for values between 10% and 90%. In October 2011, pretreatment live height was measured to the nearest centimeter using a height pole. At that time, BSD at groundline and diameter at breast height (DBH; e.g., 137 cm from groundline), were measured to the nearest millimeter. In order to quantify stem dieback or growth, stem live height was measured again 12 and 24 MAT. Live DBH was measured at 24 MAT.

Statistical Analysis

Crown Reduction

Because crown reduction response values were primarily near the fixed limits of 0 and 100%, homogeneity of variances and normality could not be achieved by standard data transformations (arcsine, arcsine square root, and log). As a result, traditional parametric and nonparametric tests could not be used to determine whether differences among treatment groups existed. Instead, Welch's ANOVA on ranked data was performed using the PROC GLM procedure in SAS (SAS 2015) to determine whether differences existed between treatments. The Ryan-Einot-Gabriel-Welsch multiple range test (REGWQ) was performed to compare treatment means at $\alpha = 0.05$. These procedures are recommended for analysis of data that are simultaneously heteroscedastic and non-normal, and they have been shown to provide good power and acceptable control for type I error rates (Cribbie et al. 2007).

Stem Live Height and Diameter

The PROC GLM procedure (SAS 2015) was used to determine differences among treatment groups for the change from pretreatment stem live height at 12 and 24 MAT and for change in DBH at 24 MAT. Tukey's HSD test was used to compare means at $\alpha = 0.05$.

Results and Discussion

Analysis for all parameters indicated a significant site and site by treatment interaction. As a result, data were analyzed and presented by site.

Crown Reduction

Welch's ANOVA revealed a highly significant ($P < 0.0001$) effect of basal stem treatments on crown reduction at 2 and 6 MAT for both sites, and at 12 MAT on the non-eroded site. Except for triclopyr ester at the 2 MAT assessment at the non-eroded site, all treatments resulted in greater crown reduction than the nontreated check (Table 2). At both sites crown reduction developed quickly with AMCP and more slowly with imazapyr and triclopyr. By 6 MAT, 100% crown reduction was observed for nearly all trees treated with AMCP, regardless of rate, tree size, or site. The lowest rate of AMCP resulted in less crown reduction (82%) compared to the higher rates at 2 MAT on the non-eroded site, which had larger trees. Standard herbicide treatments resulted in less crown reduction than AMCP treatments, except at 6 and 12 MAT on the eroded site. There, the population consisted of small, less vigorous trees that were perhaps more susceptible to herbicides. Mean crown reduction for imazapyr-treated trees changed from 62% to 100% on the eroded site and from 42% to 91% on the non-eroded site between 2 and 12 MAT, indicating that symptoms for imazapyr injury are slow to develop, as is commonly reported for herbicides with a mode of action that inhibits amino acid synthesis (Gunsolus and Curran 1999). Likewise, symptoms were slow to appear when triclopyr ester was used, with mean crown reduction increasing from 26% to 97% on the eroded site and 4% to 86% on the non-eroded site between 2 and 12 MAT.

Stem Live Height

The effect of basal stem treatment on stem live height change at 12 and 24 MAT was highly significant ($P < 0.0001$). All treatments resulted in greater stem live height reduction compared to that of the nontreated check. Nontreated checks increased in live height by 695 cm on the eroded site and 762 cm on the non-eroded site by 24 MAT (Table 3). On the eroded site, there was no difference in stem live height reduction between any of the AMCP and standard treatments at any assessment. On the non-eroded site, all AMCP treatments resulted in greater stem live height reduction than triclopyr ester but these reductions were not different from those at 24 MAT when imazapyr was used. Comparison of stem live height reduction from 12 to 24 MAT revealed that trees treated with triclopyr ester recovered 23 to 44 cm in height on the eroded and non-eroded sites, respectively.

Stem Diameter

Similar to stem live height and crown reduction, the overall effect of basal stem treatment on stem diameter at 24 MAT was highly significant ($P < 0.0001$). All herbicide treatments resulted in greater stem diameter reduction compared to that of the

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Table 2. Periodic crown reduction of *Eucalyptus benthamii* assessed 2, 6, and 12 mo after treatment following basal stem applications at an eroded and non-eroded site.[a]

		Crown reduction[b]					
		Eroded			Non-eroded		
Herbicide	Rate	2 MAT	6 MAT	12 MAT	2 MAT	6 MAT	12 MAT
	% vol/vol	%					
AMCP b	5	86 a	100 a	100 a	82 b	97 a	99 a
AMCP	10	98 a	100 a	100 a	98 a	100 a	100 a
AMCP	20	100 a	100 a	100 a	99 a	100 a	100 a
AMCP	40	100 a	100 a	100 a	99 a	100 a	100 a
Imazapyr	28.1	62 b	98 a	100 a	42 c	83 b	91 b
Triclopyr ester	75	26 c	92 a	97 a	4 d	73 b	86 b
Nontreated check	–	1 d	1 b	0 b	3 d	3 c	3 c

[a]Abbreviations: AMCP, aminocyclopyrachlor; MAT, months after treatment.
[b]Treatment means within a column followed by the same letter are not significantly different according to the Ryan-Einot-Gabriel-Welsch multiple range test on ranked data at $\alpha = 0.05$.

Table 3. Responses to basal stem herbicide application showing the change from pretreatment stem live height and diameter at breast height of *Eucaluptus benthamii* for assessments at 12 and 24 mo after treatment.[a]

		Stem height change[b]				DBH Change[b]	
		Eroded		Non-eroded		Eroded	Non-eroded
Herbicide	Rate	12 MAT	24 MAT	12 MAT	24 MAT	24 MAT	24 MAT
	% vol/vol	cm					
AMCP[b]	5	−402 a	−402 a	−586 ab	−577 a	−3.1 a	−4.2 a
AMCP	10	−430 a	−430 a	−601 ab	−601 a	−3.2 a	−4.8 a
AMCP	20	−422 a	−422 a	−638 a	−638 a	−3.0 a	−5.0 a
AMCP	40	−403 a	−403 a	−596 ab	−594 a	−3.1 a	−4.5 a
Imazapyr	28.1	−363 a	−378 a	−410 bc	−480 ab	−3.0 a	−3.2 ab
Triclopyr ester	75	−322 a	−299 a	−289 c	−245 b	−2.1 a	−1.5 b
Nontreated check	–	336 b	695 b	403 d	762 c	6.5 b	6.4 c

[a]Abbreviations: AMCP, aminocyclopyrachlor; MAT, months after treatment.
[b]Treatment means within a column followed by the same letter are not significantly different according to Tukey's HSD at $\alpha = 0.05$.

nontreated check, which increased by 6.5 and 6.4 cm at the eroded and non-eroded sites, respectively (Table 3). Stem diameter reduction at 24 MAT did not differ among herbicide treatments at the eroded site. At the non-eroded site, all AMCP treatments resulted in greater stem diameter reduction than triclopyr ester, but not imazapyr.

Triclopyr ester was consistently the least effective herbicide at both locations by all metrics (crown reduction, stem live height, and diameter), and an increase in stem live height from 12 to 24 MAT indicates less effective herbicide control from triclopyr ester treatments. These findings are in agreement with those reported by Enloe et al. (2015) that greater control of the invasive Chinese tallow was achieved with AMCP (12 or 24 g L^{-1}) compared to triclopyr ester (48 or 96 g L^{-1}) applied in an oil carrier for basal stem treatment. The effectiveness of AMCP treatments observed in our studies is also consistent with the results reported by Wilson et al. (2011) that AMCP when applied as a basal stem treatment offered effective control of other fast-growing woody plants such as Russian olive (*Elaeagnus angustifolia* L.) and salt cedar (*Tamarix* sp.). Edwards and Beck (2011) also reported similar results for Russian olive, although their trials were notably different because they applied approximately 30 ml per 2.5 cm of stem diameter, whereas 5 ml per 2.5 cm BSD was applied in our study. Although a higher herbicide rate may be required to control Russian olive compared to that for eucalyptus, it appears that other studies have generally not investigated the lowest rates necessary to achieve control of some woody plants. Future studies of woody plant control with AMCP should examine lower rates than used in our study.

Although these studies tested AMCP rate response and the effects of tree size in comparison to the efficacy of standard herbicides, the potentially important effects of application timing and eucalyptus species susceptibility were not determined. This would require studies across multiple seasons for those eucalyptus species most common in plantations globally, such as *E. camaldulensis* Dehnh., *E. grandis* W. Hill ex Maid., *E. urograndis* (hybrid of *E. grandis* and *E. urophylla* S.T. Blake), *E. amplifolia* Naudin, and others. Future research regarding species susceptibility could look to the work by Morze (1971) that identified susceptible and resistant eucalyptus species for picloram. The highly effective control of *E. benthamii* in this study represents significant progress in the development of effective herbicide prescriptions for eucalyptus management and indicates that future research with different species and timings would be worthwhile.

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