

# Stocktype and grass suppression accelerate the restoration trajectory of *Acacia koa* in Hawaiian montane ecosystems

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**Abstract** Restoring degraded mesic-montane forests represents a major challenge in maintaining functioning ecosystems throughout the tropics. A key example of this lies in Hawai'i, where restoring native koa (*Acacia koa*, A. Gray) forests are a top conservation and forestry priority because of the critical habitat and high-value timber products that they provide. Efforts to restore koa forests, however, are directly impeded by extensive, non-native kikuyu grass (*Pennisetum clandestinum* Hochst. ex Chiov.) swards occupying deforested montane landscapes. In this study, we implemented a combination of grass suppression and selection of koa seedling stocktypes to measure outplanting performance in a naturalized site on the island of Maui. Seedlings were grown in a nursery in two root container sizes (111 and 207 cm<sup>3</sup>) and subsequently outplanted into grass-dominated plots that were either untreated or suppressed with a high-rate herbicide combination of imazapyr and glyphosate (1.7 kg a.i. ha<sup>-1</sup>, respectively), 30 days prior to planting. Across all treatments, seedling survival was high (>95 %). Thirty months after planting, trees from the larger stocktype had significantly greater growth in height and root-collar diameter. Initial grass suppression resulted in trees that were 34 % taller with 66 % larger root-collar diameters after 30 months. Herbicide treated plots also had significantly higher leaf area indices (2.6 vs. 1.8 m<sup>2</sup> m<sup>-2</sup>), indicative of higher photosynthetic capacity and canopy closure. Grass suppression increased soil temperature along with soil moisture in the first year followed by a dramatic drop in moisture corresponding to large growth

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responses by koa seedlings after the first year. These results demonstrate how the combination of fundamental silvicultural practices in the nursery and on the outplanting site can accelerate tree growth to meet restoration goals in shorter time intervals. This is a first report of koa (a leguminous species) tolerance to a high-rate, pre-plant application of the herbicide active ingredient imazapyr.

**Keywords** Hawai'i · Seedlings · Stocktype · Site preparation · Grass suppression · Soil moisture

## Introduction

Koa (*Acacia koa* A. Gray) is an endemic canopy species dominant in the mesic-montane forests types of Hawai'i. Unfortunately, these forests have been degraded over the last two centuries by anthropogenic activities, specifically, degradation through deforestation for conversion to pasture (Wagner et al. 1990). This is particularly troubling because koa serves as critical habitat to several endemic fauna, including the 'Akiapola'au (*Hemignathus munroi* Pratt, 1979), one of the Hawaiian Honeycreepers, and many of its congeners (Menard 2001; Pejchar et al. 2005). Fortunately, koa is also one of the most economically valuable timber species in the world (Elevitch et al. 2006), thereby creating unique potentials for incentivizing koa habitat restoration as multi-purpose systems (Pejchar and Press 2006). Interest is therefore increasing among government agencies and private landowners in restoring the Hawai'i's native forests. Yet despite the possible economic, ecological, and cultural values of koa forests, not a single stand of koa has been through a full silvicultural rotation in Hawai'i (Baker et al. 2009). If any intrinsic gains are to be made from restoring koa forests, it follows that the quickest route to achieve this goal should be implemented.

Advances in many aspects of forest restoration practices have helped to improve koa forest establishment in recent years. More specifically, techniques in breaking seed dormancy (Yoshinaga 1998), seedling outplanting (Scowcroft and Jeffrey 1999; Denslow et al. 2006), site preparation (Scowcroft and Adee 1991), and stocking guidelines (Baker and Scowcroft 2005; Scowcroft 2012) have been highlighted as evidenced by literature. Furthermore, improved nursery practices have demonstrated how seedling quality translates into better establishment and performance in the field (Davis et al. 2011; Dumroese et al. 2009, 2011). Once planted into the natural environment, a sufficient level of root egress (specific to site limitations) is required for a seedling to maintain a proper balance of photosynthetic carbon assimilation with compensation for stomatal water losses and increased nutrient demand (Burdett 1990; Grossnickle 2005). Once a seedling is conditioned to the hydrological cycle on a site, impediments to survival and growth are greatly reduced (Grossnickle 2012).

Various types of site preparation can alleviate harsh conditions (Rose et al. 1999), but proper stocktype selection can also do the same (Chirino et al. 2008; Villar-Salvador et al. 2012). The mesic, montane zones suitable to koa restoration in Hawai'i are largely dominated by non-native, stoloniferous kikuyu grass (*Pennisetum clandestinum* Hochst. ex Chiov.) stands; this grass forms large uninterrupted mats, effectively inhibiting natural tree regeneration (Shlisky 2000). On sites with heavy grass competition, Scowcroft and Adee (1991) found that a high rate of the herbicide glyphosate (e.g., 6.07 kg a.i. ha<sup>-1</sup>) was more

effective than scarification in reducing vegetation competition. In trials simulating extreme moisture deficit, Aghai et al. (2014) found that larger western larch (*Larix occidentalis* Nutt.) stocktypes facilitated greater root egress following outplanting. Additionally, Pinto et al. (2012) found that increasing seedling root length of *Pinus ponderosa* by as little as 5 cm (i.e., via larger stocktype) was critical to survival in soil conditions that rapidly dried soon after planting.

Whether the goals are to promote ecological services or commercial products, seedling survival is of primary importance to reforestation efforts (Scowcroft and Jeffrey 1999). A secondary consideration is growth rate of established plantations. Optimizing productivity to achieve the aforementioned goals in the shortest time interval has not been thoroughly evaluated for koa across all suitable island biomes. Therefore, it may be possible with this species, to reduce the amount of time ecosystem services may be realized. For instance, a 15 year-old “scarified” koa plantation was comparable to an “old growth” koa forest in suitability as a foraging habitat for the Hawaiian honeycreeper demonstrating the opportunity of commercial forestry in provisioning natural capital (Pejchar et al. 2005; Pejchar 2007). This is an increasingly important consideration as the legacy supply of mature, large canopy koa is virtually depleted, and many landowners are now seeking koa reforestation as a viable land-use alternative to pasture-based agriculture (Dudley 1997). Further, Loudat and Kanter (1996) have speculated on 20-year harvest rotations with the assumption of meeting economically viable maturity and size class benchmarks. However, extensive knowledge gaps remain in koa growth projections to maturity, prohibiting justified investments (Grace 1995).

Reforestation using nursery-produced seedlings is often the quickest, most reliable way to ensure successful establishment and rapid growth following outplanting. The silvicultural combinations of stocktype selection and effective site preparation are just two factors that contribute towards reforestation success and are likely treatments to increase the speed of koa reforestation. The objectives of this study were therefore aimed at characterizing post-planting survival and early growth responses of outplanted koa resulting from a combination of silvicultural practices including the influence of: (a) nursery container volume and (b) pre-plant grass suppression. We suspect that the combination of larger container sizes and effective grass suppression would significantly complement higher seedling survival and greater early (outplanted) growth performance, to demonstrate an accelerated approach to achieve koa-based ecosystem services and provisions.

## Materials and methods

### Nursery culture

Koa seeds were sourced from a naturalized, open-pollinated population on Maui (courtesy of Nicklos Dudley, Hawai'i Agriculture Research Center). Seeds were manually scarified using nail clippers to incise the seed coat, then imbibed in a 24-h bath of aerated deionized water and subsequently sown across a single tray of peat:vermiculite (1:1) medium (Luna et al. 2008). When radical lengths reached 0.5 cm (or longer), germinants were transplanted into modified Jiffy<sup>®</sup> containers (one seed per cell). Sown containers were covered with Deluxe Seed Guard<sup>™</sup> germination fabric (DeWitt Company Inc., Sikeston, Missouri) and mist irrigated twice daily until true leaf epicotyls were present.

Seedlings were grown in a greenhouse at the University of Idaho Pitkin Forest Nursery in Moscow, Idaho (46°43'N, 117°00'W) using modified Jiffy® compressed peat pellet containers (Jiffy Products of America, Inc., Norwalk, OH). Fully hydrated and expanded (13 cm height × 5 cm diameter), Jiffy® 50150 pellets were adjusted to plug depths of 13, or 7 cm, resulting in two experimental volumes: 207, and 111 ml, respectively ( $n = 135$  containers per volume). Hereafter, 13 and 7 cm stocktypes will be referred to as LS or SS, respectively. Following adjustments, the containers were placed in Jiffy® air trays (6 × 6 configuration).

Sowing was completed by 20 March 2011, with nutrient fertilization beginning 2 weeks later. Nutrient concentration of irrigation water was tailored to provide the identical targets to the two differing stock sizes (Aghai et al. 2014), and irrigation rates were determined using gravimetric water content (Dumroese et al. 2015) to mitigate experimental confounding. Using an overhead irrigation system, Peters® Professional Conifer Grower™, along with calcium nitrate (The Scotts Company, Marysville, Ohio) and S.T.E.M. (Soluble Trace Element Mix) were applied to seedlings twice a week for 8 weeks at 100 ppm N. Seedlings were transported to Ulupalakua Ranch Nursery (Maui, HI) and allowed to acclimate for approximately 30 days. Following transport, seedlings were placed in subirrigation trays to irrigate and apply four weekly applications of Peters® Professional Conifer Finisher™ at a rate of 25 ppm N. Seedlings were selected for uniformity within root container sizes on the day to be outplanted on 30 June 2011.

## Field site and study layout

The field site is located on Ulupalakua Ranch property (20°40'06 N, 156°20'59 W) on the island of Maui. Positioned on the southwestern rift zone of Haleakalā Mountain, the field site has a west aspect and is 1455 m, a.s.l. The site is a slight depression with a <10° slope. Soils are a volcanic ash of the Kaipoi series (classified as medial, amorphic, isomesic Humic Haplustands); characteristics are deep, well drained soils consisting of loam texture with a high CEC and neutral to acidic pH (NRCS 2015; CTAHR 2015). Precipitation records from the past two decades average 850 mm annually, 158 mm in January (most), and 20 mm in July (least) (Giambelluca et al. 2013). Thirty year records indicate an average temperature of 14.3 °C; the coldest month is Feb with an average of 12.4 °C, high of 21.8 °C, and low of 0.0 °C; the hottest month is Aug with an average of 16.3 °C, high of 25.8 °C, and low of 5.4 °C (Giambelluca et al. 2014). This site is characterized as a Temperate Subhumid micro-climate that was formerly 'ōhi'a (*Metrosideros polymorpha* Gaudich.) and *Acacia koa* cloud forests, but now consisting primarily of introduced kikuyu grass along with minor representations of C3 grasses including Yorkshire fog (*Holcus lanatus* L.) and sweet vernal grass (*Anthoxanthum odoratum* L.). The experimental site was fenced to keep grazing cattle out. Within this enclosure, a smaller area was divided into four ~150 m<sup>2</sup> treatment blocks, each containing a replicate of herbicide-treated and untreated grass plots (~40 m<sup>2</sup>).

## Site preparation and planting

On 30 May 2011, two grass suppression treatments, an herbicide treatment (hereafter referred to as herbicide or H+) and an untreated control (hereafter referred to as no herbicide or H-), were applied to each of the four blocks. A combination of the active ingredients glyphosate (Ranger® Pro, EPA reg. no. 524-517, Monsanto Company, St. Louis, MO) and imazapyr (Polaris AC, EPA reg. no. 228-570, NuFarm Americas Inc., Burr Ridge, IL) were applied at 1.7 kg a.e. ha<sup>-1</sup>, respectively, with a total volume rate of

250 L ha<sup>-1</sup>. A commercial blended methylated seed oil and organosilicone surfactant adjuvant was added to the final spray formulation at 1 % v/v. Herbicide was applied by hand using a handheld XP Boomjet® 10L (TeeJet®, Wheaton, IL) wand calibrated to deliver a 4 m coarse droplet swath at 3 L min<sup>-1</sup> and 307 kPa. Seedlings were ~105 days old when outplanted into the study site on 30 June 2011 (30 days after herbicide treatment). Outplanting operations were conducted over a 48-h time period. To avoid planter technique confounding, all seedlings were hand-planted by block via two individuals, with each planter planting half of all the stocktypes. Each container size was planted in rows randomly assigned within the main treatment plots with two other non-experimental rows in close 1 × 1 m spacing with 20 experimental seedlings and a total of 40 seedlings. Each spray plot had a 1 m suppression buffer surrounding the tree planting area.

## Data sampling

Prior to outplanting, subsets of seedlings were destructively harvested to measure seedling biomass and root morphology. Shoot and root portions were partitioned at the root-collar and dried at 60 °C for 72 h to a stable dry weight; shoot dry weight (SDW) and root dry weight (RDW) were subsequently recorded. Root volume (RV) was determined by water displacement (Burdett 1979; Harrington et al. 1994), and root tips were counted. Immediately following outplanting, seedling height and root-collar diameter (RCD) were measured. Periodic growth assessments were made at 4, 8, 12, and 30 months after planting. Heights were determined as a measure from ground-line to the tallest apical meristem; RCDs were measured approximately 1 cm above ground-line. Height and RCD growth were calculated as the current period's measure minus the outplanted measurement. On the final growth assessment, 30 months after planting, leaf area index was measured for each treatment × block combination (n = 8) using a sunfleck ceptometer (model SF-80, Decagon Devices, Inc., Pullman WA).

Soil edaphic measurements were collected within each treatment (n ≥ 5, due to some sensor malfunctioning) including: volumetric water content ( $\theta$ ) and soil temperature at 10 and 25 cm depths. Measurements were collected hourly, in situ, using ECH<sub>2</sub>O EC-TE soil moisture sensors connected to Em50 data loggers (Decagon Devices, Inc., Pullman, WA). MPS-1 sensors (also from Decagon Devices) were used to collect matric soil water potential ( $\Psi_m$ ) hourly, in situ, at a depth of 10 cm.

Analysis of variance (PROC GLIMMIX) was used to examine the fixed effects of container depth on greenhouse-grown *Acacia koa* seedling height, RCD, SDW, RDW, total RV, number of root tips, and root-to-shoot ratio (R:S), prior to planting; block was classified as the random variable. The field experiment was analyzed as a repeated measure, split-plot analysis of variance (PROC MIXED) with spray treatments as the whole-plot and root container size (x = 8) as the split-plot, blocked four times to examine the fixed effects of container type and herbicide treatment on *Acacia koa* seedling height, RCD, and survival; block was classified as the random variable. All analyses were performed using SAS (SAS 9.4, SAS Institute Inc., Cary, NC, USA;  $\alpha = 0.05$ ).

## Results

After 10 weeks in nursery production (~2 months before planting), stocktype showed significant influence on seedling morphology (Table 1). Height and RCD were both significantly greater for LS seedlings ( $P = 0.0021$ ). Shoot biomass significantly increased

**Table 1** Morphological characteristics of *Acacia koa* seedlings after 10 weeks of nursery culture

Container depth/volume (cm/ml)	Height (cm)	Root-collar diameter (mm)	Shoot dry mass (g)	Root dry mass (g)	Root volume (ml)	# Root tips
7/111	32.7 (0.9)a	3.7 (0.1)a	6.72 (0.25)a	2.11 (0.10)a	4.27 (0.22)a	1150 (108)a
13/207	36.3 (0.7)b	4.2 (0.1)b	9.82 (0.48)b	3.05 (0.15)b	6.80 (0.42)b	2019 (129)b

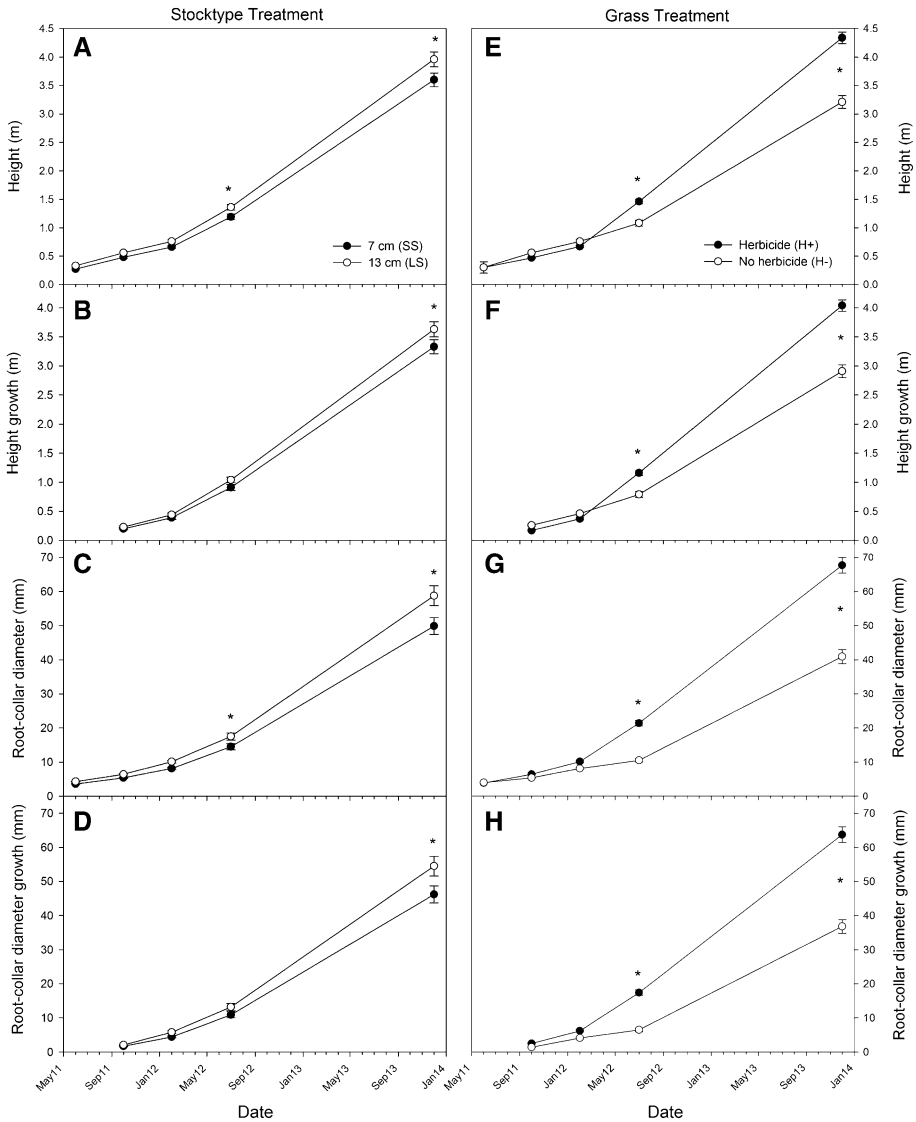
Root-collar diameter is the main seedling stem measured approximately 1 cm above ground-line. Standard errors are in parentheses. Columns with the same letter are not significantly different ( $P < 0.05$ )

with stocktype ( $P < 0.0001$ ). Seedling RV, number of root tips, and root biomass were lower for SS versus LS ( $P < 0.0001$ ). R:S was not different ( $P = 0.3559$ ) between seedling stocktypes (R:S = 0.33, SE = 0.017).

Post-outplanting, analysis showed some morphological changes over time (Fig. 1). No significant stocktype  $\times$  herbicide treatment or stocktype  $\times$  herbicide  $\times$  time interactions for height, height growth, RCD, and RCD growth were detected; however, a stocktype  $\times$  time interaction was significant for the same variables ( $P < 0.0445$ ). In general, most morphological and growth characteristics increased over time ( $P < 0.0423$ ), and stocktype differences were not evident in the first 8 months of the outplanting experiment. At 12 months, LS stocktypes became significantly taller and had larger RCDs ( $P < 0.0228$ ), while growth characteristic differences were not evident ( $P > 0.1031$ ). After 30 months, LS stocktypes were significantly taller, had greater height growth, had larger RCDs, and greater RCD growth ( $P < 0.0001$ ; Fig. 1A–D).

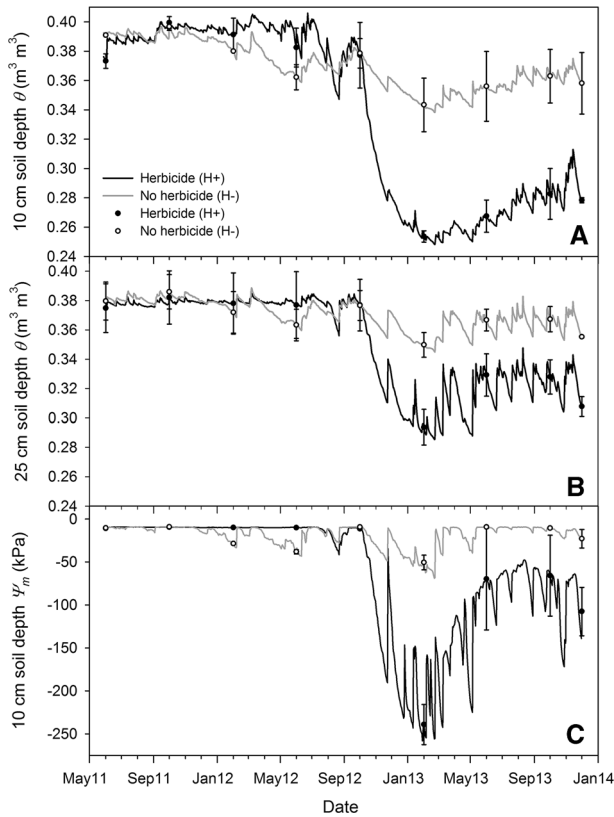
Outplanting survival was high (>95 %) throughout the entire experiment. Mean survival was slightly higher in the H+ treatment plots, although not significant ( $P = 0.131$ ). Post-outplanting analysis showed an herbicide treatment  $\times$  time interaction ( $P < 0.0001$ ) for all morphological and growth characteristics (height, height growth, RCD, and RCD growth). As previously mentioned, these variables significantly increased with time, but H+ treatment height and RCD characteristics only began to significantly outperform H– at 12 months. These differences were maintained until the end of the outplanting measurements ( $P < 0.0001$ ; Fig. 1E–H). Along with the larger seedling size at 30 months, H+ also measured significantly higher leaf area index (2.6 vs. 1.8 m<sup>2</sup> m<sup>-2</sup>;  $P = 0.0288$ ).

Over the course of the study, mean  $\theta$  and  $\Psi_m$  varied greatly. In the first 16 months, values for each soil moisture parameter diverged only slightly with periodic drying between precipitation events. During this time, 10 cm  $\theta$  H+ and H– averaged 0.39 (SE = 0.009) and 0.38 m<sup>3</sup> m<sup>-3</sup> (SE = 0.004), respectively. Within the first 30 days after herbicide application, mean  $\theta$  values at both the 10 and 25 cm depths were similar, only varying slightly with precipitation. About 180 days after grass suppression treatment, mean values began to separate slightly with 10 cm H– drying faster and more between wetting cycles (Fig. 2A, B). The same trend was true for  $\Psi_m$ ; however, H+ maintained fairly steady, less negative values, while H– clearly illustrated reduced water availability (Fig. 2C). In this same 16 month period,  $\Psi_m$  for H+ and H– averaged –11 kPa (SE = 0.8) and –15 kPa (SE = 1.2), respectively. Beginning in October 2012,  $\theta$  and  $\Psi_m$  dry down trends reversed with respect to treatments. After a brief wet period from late August to September 2012, where  $\theta$  and  $\Psi_m$  values seemed to equilibrate, moisture depletions were observed in each treatment but were much more dramatic in H+ (Fig. 2A–C). This trend remained through to the last morphological assessment at 30 month. During this time (17–30 months), the



**Fig. 1** Height and root-collar diameter characteristics for *Acacia koa* seedlings measured over a 30 month period. Stocktype treatments are for 111 and 207 ml sized containers designated by 7 cm (SS) and 13 cm (LS), respectively. Significant results are indicated by asterisk and are at the  $\alpha = 0.05$  level

average 10 cm  $\theta$  for H+ and H- were 0.28 (SE = 0.011) and 0.36  $\text{m}^3 \text{m}^{-3}$  (SE = 0.019), respectively. Mean  $\Psi_m$  values were also much lower for H+ at this time (-120 [SE = 39.5] and -22 kPa [SE = 3.6] for H+ and H-, respectively). Over the course of the entire study period (30 months), the 10 cm H+ reached the lowest  $\theta$  value of 0.25  $\text{m}^3 \text{m}^{-3}$  while H- reached 0.34  $\text{m}^3 \text{m}^{-3}$ . Average  $\Psi_m$  values for H+ and H- treatments were -61 (SE = 18.7) and -18 kPa, (SE = 2.3), respectively. H+ treatments reached a minimum of -259 kPa while H- treatments reached -79 kPa.



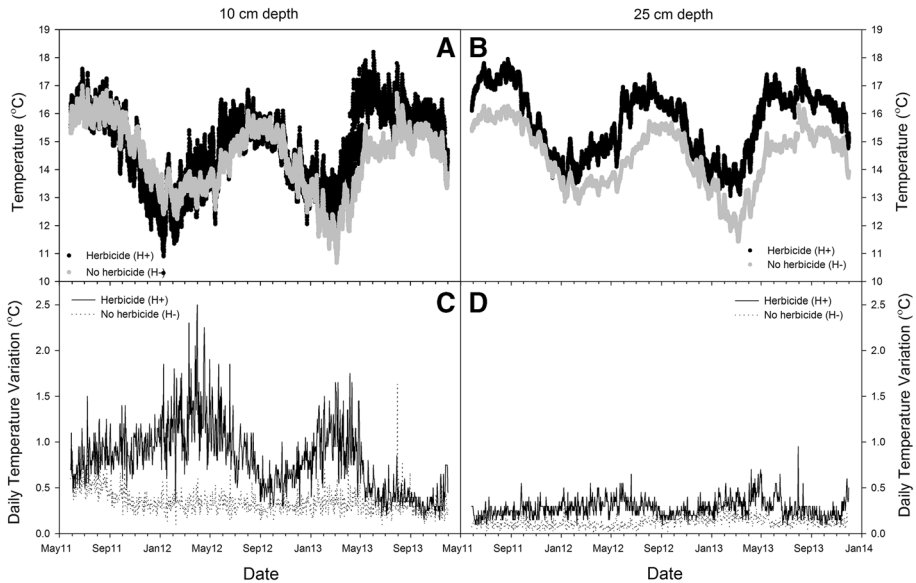
**Fig. 2** Seasonal soil moisture trends of an *Acacia koa* seedling outplanting site. Experimental plots were either treated with herbicide (H+; glyphosate + imazapyr) or no herbicide (H–; vegetation intact). Data was logged hourly for 30 months. Because of the large amount of data presented, standard errors are shown as points in 4 month intervals ( $n \geq 5$ )

Soil temperatures showed seasonality with lows during the winter months (January through February) and highs peaking in the summer months (July through August). At the 10 cm depth, average temperatures were 14.9 (SE = 0.17) and 14.4 °C (SE = 0.18) for H+ and H– treatments, respectively. Of the two treatments, highest and lowest temperatures were reached in H+ (18.2 and 10.3 °C, correspondingly). Temperature data showed mean daily variation values of 0.82 (SE = 0.17) and 0.35 °C (SE = 0.03) for H+ and H– plots, respectively. The most variation was observed in H+ at 2.5 °C, compared with 1.6 °C in H–. Mean temperatures at 25 cm trended warmer throughout the study period for the H+; mean temperature variation was twice as high in H+ than H–, 0.28 (SE = 0.10) and 0.12 (SE = 0.04) respectively (Fig. 3B, D).

## Discussion

Initial seedling establishment and survival on a site can depend on many factors (Grossnickle 2012). The high seedling survival (>95 %) measured in this study, in spite of grass competition and herbicide injury imposed by the respective H– and H+ treatments, is





**Fig. 3** Seasonal trends in soil temperature at two depths, 10 and 25 cm ( $n \geq 5$ ). Experimental plots were either treated with herbicide (H+; glyphosate + imazapyr) or no herbicide (H–; vegetation intact). Daily temperature variation was calculated as  $T_v = T_{\max} - T_{\min}$  for a 24 h period (C, D)

likely attributed to the quality assurance and quality control of stocktypes prepared in the nursery. Survival is a necessity for all restoration projects and in many cases may be the only measure of success for projects with more esoteric, long-term outcomes and no inherent time constraints. However, Pejchar et al. (2005) show how ecosystem provisions and services can be delivered anthropogenically (e.g., soil scarification) in rapid succession (i.e., 15 years), which is the impetus of this study. Ultimately, delays in establishing functionally sustainable ecosystem services are equivalent to lost opportunities, where silvicultural inputs can compensate by reducing this timeline with greater rates of productivity.

Historically, koa reforestation projects in Hawai'i have been installed using stocktypes that are small in size (e.g., 50 cm<sup>3</sup>) (Scowcroft and Adee 1991; Whitesell 1990). Dumroese et al. (2011), however, demonstrated how larger koa stocktypes (from 164 to 656 cm<sup>3</sup>) could be produced in the nursery with comparable quality to the smaller, 50 cm<sup>3</sup> standard stocktypes. We chose our stocktypes because they offer several benefits. The first addresses the confounding variables of container density and volume in stocktype testing (as advised by Pinto et al. 2011). The second relates to the potential benefits of larger seedling size and seedling quality characteristics as observed from Dumroese et al. (2011) but without producing a seedling that might be considered too large or too expensive for current reforestation operations in Hawai'i (i.e., the 656 cm<sup>3</sup> size used by Dumroese et al. 2011). The larger stocktypes in this study could have contributed to the high survival described above. Previous research has shown that larger stock types have performed better in poor soil environments (e.g., extreme periodic drought) (Villar-Salvador et al. 2012; Pinto et al. 2011). Other koa field trials have reported much lower seedling survival rates after 1 year, particularly in the absence of site preparation (Davis et al. 2011; Scowcroft and Adee 1991). In the course of this study, the LS stocktype (207 cm<sup>3</sup>) grew larger than the

SS (111 cm<sup>3</sup>). Our findings build upon existing research that validates the utility and performance of larger stocktypes with improved survival and growth performance (Villar-Salvador et al. 2012; Pinto et al. 2012; Aghai et al. 2014).

Our study shows that effective site preparation with a short- and long-term grass suppression tactic (e.g., glyphosate + imazapyr) can increase growth and canopy closure with koa in Hawai'i. Scowcroft and Adee's (1991) early work with chemical and mechanical grass suppression (i.e., short term), in part supports this. Thaxton et al. (2012) also demonstrated that seedling performance for several Hawaiian native plant species was improved when grass was physically removed. These studies have both posited that a potential reduction in moisture competition could have been the driver for improved seedling performance, regardless of the method of grass suppression. Other studies found that smaller seedling stocktypes (e.g., conifer species) demonstrated exceptional survival and growth, but only when the combination of seedling quality and proper site preparation were factors accounted for (Aghai et al. 2014; Chirino et al. 2008; Oliet et al. 2012). In our study, grass suppression resulted in greater volumetric water content ( $\theta$ ) observed within 4 months of herbicide suppression and extending to 12 months after planting, which coincides with the time when koa in the H+ plots become significantly larger, as described below.

Koa seedlings from the H+ plots were stunted in the first 8 months after planting, with classical symptoms of imazapyr injury including, arrested apical growth and proliferation of lateral branching (personal observation by J.J.K. Leary). However, no apparent lethality was observed as a result of the herbicide application. It is worth noting that imazapyr was applied at the maximum allowable rate (i.e., 1.7 kg a.e. ha<sup>-1</sup>) only 30 days prior to out-planting. Imazapyr is known for having soil persistence with a degradation half-life of 25–142 days and able to provide suppression from 3 months, up to 2 years, depending on the environment (Anonymous 2007). This is a first report of *Acacia koa* exhibiting exceptional tolerance to a high rate of imazapyr in a pre-plant application where the herbicide was possibly taken up via root absorption. The tolerance mechanism is likely a metabolic process of hydroxylating the imidazolinone ring structure (Anonymous 2007). In fact, at 12 months after planting, these stunted koa saplings in the H+ plots superseded growth rates of the saplings in H– plots, clearly showing the ameliorative effects against competition, despite the early set back.

RCD is often most correlated with desirable seedling attributes such as increased root size and hydraulic transport, which often confer higher survival and growth (Grossnickle 2012). It is possible that despite the mild inhibition in above ground growth, grass suppression may have facilitated an opportunity for koa seedlings to maintain hydraulic systems and some root growth. This in turn translated into rapid height growth once toxicity subsided. The resultant greater height and canopy growth likely equated to greater potential transpiration thereby depleting soil moisture in these plots, especially in the absence of precipitation inputs. This is further supported by the dramatic, negative  $\theta$  and  $\Psi_m$  trends observed after 12 months correlating to koa growth and demand.

The dramatic changes measured in the below ground environment (e.g., temperature and moisture) are strong indicators of koa growth measured over time. However, there are likely other (unmeasured) factors that have been modified by these same treatments, further contributing to koa health and productivity. For instance, koa is a symbiotic partner with a wide range of soil microbes, including the N<sub>2</sub>-fixing bradyrhizobia and arbuscular mycorrhizal fungi (AMF) (Leary et al. 2006; Miyasaka and Habte 2001). For an arboreal species, koa has a rather shallow root system (Whitesell 1964). At 30 months after planting, all H+ plots were observed to have extensive koa root networks occupying the

soil surface that were also profusely nodulated. Meanwhile, all H— plots were extensively occupied by grass root networks where nodulated koa root systems were not readily observed. We were not trained to observe AMF, but koa is known to be moderately dependent on AMF symbiosis in soils with low available phosphorous, as the soils of this study (CTAHR 2015). On the other hand, no literature was found on the host status of kikuyu grass, but it is likely to be a less obligate non-host to AMF due to its fine root network (Miyasaka and Habte 2001). The seedlings were not pre-inoculated in the nursery, so it is most likely that these symbiotic interactions occurred with the indigenous microbial populations on the site. It is plausible that a shift to a rhizosphere dominated by koa would trigger population increases of these indigenous microbial symbionts previously surviving as latent, free-living organisms.

This experiment demonstrates that kikuyu grass, as a naturalized occupant of montane landscapes in Hawaii, can impose significant impediments on koa restoration productivity. However, this study also further validates the combination of basic silvicultural practices to ameliorate these competitive conditions and accelerate the establishment of potential ecosystem services accommodated by young koa habitat. Further research is warranted in many areas towards optimizing strategies and tactics in establishing koa-based ecosystem services: (1) Expanded trials on herbicide-based site preparation that investigates timing and rate combinations, (2) Expanded trials investigating the effects of stocktype sizing in combination with planting density, (3) A study of the soil microbiology as indicators of a transitioning landscape, (4) Cost-benefit analyses that matches inputs to expected returns in ecosystem services and (5) Development of a monitoring remote sensing platform that correlates microclimatic data with koa growth productivity and ecosystem provisioning.

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