

Evaluating the Fecundity of *Rhamnus frangula* (Glossy Buckthorn) Cultivars through Seed Yield, Seed Size, and Germination

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ABSTRACT. Invasive plants are nonnative to a region and can outcompete native plants, causing ecological harm. *Rhamnus frangula* L. is an invasive shrub in the United States, although cultivars are still commonly used for landscaping purposes, especially in the Midwest. The fecundity of the *R. frangula* cultivars ‘Columnaris’ Tall Hedge (TH), ‘Ron Williams’ Fine Line[®] (RW), Advanced Line 1 (AL1), and Advanced Line 2 (AL2) was evaluated through fruit production, seed production, seed size, and germination rates. Fruit was collected from these cultivars planted outdoors in the ground, and seeds were cleaned from the fruits. Seed germination was conducted under intermittent mist over 9 weeks. Over the 2 years of fruit collection, the three narrow-leaved cultivars (RW, AL1, and AL2) had greatly reduced fruit and seed production compared with TH. The RW, AL1, and AL2 cultivars produced an average of 79.1, 3.63, and 0.63 fruits, respectively, and 167.0, 6.75, and 1.00 seeds, respectively, per shrub in a year. Seed size was significantly smaller for RW and AL1 compared with TH, and RW also had a lower germination rate than TH. The low seed production of the narrow-leaved cultivars compared with TH suggests that these cultivars would be a better choice for landscaping use because they are less likely to produce large seed crops which could establish invasive populations. The reduced seed size and germination rate of RW in comparison with TH further suggests a lower invasive potential, as seeds are less likely to germinate and establish. Overall, the *R. frangula* cultivars, RW, AL1, and AL2 may be less likely than TH to contribute to invasiveness.

Nonnative invasive plants can take over and cause environmental harm to a landscape, outcompeting natives and disrupting the local ecology (Simberloff et al. 2013). Invasive plants have high fecundity (Jelbert et al. 2015), vigorous and rapid growth (Graebner et al. 2012), and are adaptable to different types of habitats (Funk 2008). *Rhamnus frangula* L. (*Frangula alnus* P. Mill.), commonly known as Glossy Buckthorn, is a deciduous shrub invasive to North America. *R. frangula* was introduced to North America first to Ontario (Frappier et al. 2003) and later to the midwestern United States for wildlife habitat improvement (Webster

et al. 2006). It now inhabits eastern North America from southeastern Canada south to Tennessee (Gucker 2008).

Its original native range extends from Europe to northern Africa to western Asia (Rehder 1927). It is in the Rhamnaceae family, which also includes another invasive shrub to the United States, *R. cathartica* L. (Common Buckthorn) and a United States native shrub, *Ceanothus americanus* L. (New Jersey Tea). The habit of *R. frangula* is a tall shrub or small tree growing up to 7 m tall, and can be single or multistemmed (Gucker 2008). The leaves are ovate to obovate-oblong (Rehder 1927; Fig. 1A), and cultivars such as ‘Ron Williams’ and ‘Asplenifolia’ can have linear to linear-lanceolate leaves (Fig. 1B). The flowers are small, nonshowy, and yellow in color (Fig. 2A).

Various cultivars of *R. frangula*, including ‘Asplenifolia’ (AS), ‘Columnaris’ Tall Hedge (TH), ‘Ron Williams’ Fine Line[®] (RW), Advanced Line 1 (AL1), and Advanced Line 2 (AL2), are all targeted for landscaping purposes. The narrow-leaved cultivars (AS, AL1, AL2, and RW) all have linear to linear-lanceolate

leaves, whereas TH has ovate leaves typical of the *R. frangula* species. The narrow upright habit, dense foliage and tall habit make it an attractive option for shrub hedge rows. The narrow-leaved cultivars all have narrow upright habits, while AL1 and AL2 are shorter plants with more dense foliage.

R. frangula produces large amounts of glossy black drupe fruits that ripen from July through September (Gucker 2008; Fig. 2B). The fruits are well-liked by various species of birds (Craves 2015), and the laxative property in the fruit makes the birds an effective mode of spread for this plant (Michigan Department of Natural Resources 2012). The original spread of the species from the midwestern United States is thought to be attributed to European starlings (Howell and Blackwell 1977). Each fruit contains up to four seeds (Michigan Department of Natural Resources 2012), with an average of 1.5 seeds per fruit (Gucker 2008).

R. frangula’s invasiveness and ecological damage can be partly attributed to its vigorous growth and dense canopy cover, which can shade out understory plants and saplings (Fagan and Peart 2004; Krock and Williams 2002). *R. frangula* is also adaptable and can invade various habitats (Sukachev 1928). It especially establishes in disturbed lands, but it also thrives in and colonizes wetlands. Glossy Buckthorn invasiveness is also due to high propagule pressure from numerous seeds produced in sites that have unfavorable environmental conditions (Berg et al. 2016).

In this study, we used fruit production, seed production, seed size, and seed germination percentage as predictors of the invasive potential of four *R. frangula* cultivars. The objective of this study was to determine if there are cultivars of *R. frangula* that have reduced fecundity and are suitable for landscaping use. Similar fecundity studies have been performed on other invasive species including various species and cultivars of barberry (*Berberis* spp.; Lehrer et al. 2006), butterfly bush (*Buddleia* spp.; Anisko and Im 2001), winged euonymus (*Euonymus alatus*; Brand et al. 2012), and the *R. frangula* cultivars ‘Columnaris’ and ‘Asplenifolia’ (Wheeler and Starrett 2001). Wheeler and Starrett (2001) concluded that ‘Asplenifolia’ may have a lower invasive potential than TH due to

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Fig. 1. *Rhamnus frangula* cultivars (A) ‘Columnaris’ with ovate leaves and (B) ‘Ron Williams’ with lanceolate leaves.

its lower seed germination rate. In this study, it is hypothesized that other linear/lanceolate-leaved cultivars will show a similar reduced invasive potential compared with the broad-leaved TH cultivar.

Materials and methods

This study was performed from 2021 to 2023. Four cultivars of *R. frangula*, AL1, AL2, RW, and TH were grown in the field at the University



Fig. 2. *Rhamnus frangula* (A) small, but numerous yellow flowers and (B) glossy black fruit.

of Connecticut, Storrs, CT. Plants were installed in the field in Sep 2020 and were provided trickle irrigation as needed during the 2020, 2021, and 2022 growing seasons. Cultivar plantings (one plant per experimental unit) were arranged with four replications and in a randomized complete block design (RCBD).

Fruits were harvested in 2021 and 2022 from the plants every 2 to 4 d as they ripened from July to August, and every 7 to 10 d when fruit ripening slowed during September through November. As the fruits were harvested, they were counted, and the seeds were cleaned from the fruit with deionized water. Seeds were counted and stored in plastic sandwich bags at 38 °C until the end of November when seed collection ended.

In 2021, water washed seeds were further cleaned with a 10% bleach solution, thoroughly rinsed, and allowed to air dry. These seeds were later scarified by soaking them in 12 N sulfuric acid for 7 min, followed by repeated rinsing with water, based on the recommendations of Heit (1968) to use a 20-min acid scarification treatment. Scarified seeds were put into a moistened fine sand in sandwich bags with three times or more the amount of sand than seeds and stratified at 38 °C for 90 d. After stratification, seeds were soft and not suitable for germination because of the sulfuric acid treatment.

In 2022, seeds were cleaned with deionized water, but no bleach treatment and no acid scarification were used. Seeds were stratified as described for 2021. After stratification, seeds from TH and RW cultivars, where there were enough seeds for germination tests, were put into Connecticut flats containing ProMix FPX Bio-fungicide Fine Germination Media (Rivière-du-Loup, QC, Canada). Seeds were covered with 1 mm of media. An experimental unit was 25 seeds of one cultivar in one tray (or block). The experiment was arranged in a randomized complete block design with eight replications. Trays were put under intermittent mist in a greenhouse with set points of 21/17 °C day/night temperature thresholds, and the number of germinated seeds (as defined by a radicle emergence or seedling emerging through the media) was recorded weekly.

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Data were analyzed in SAS Version 9.4 (SAS Institute, Cary, NC, USA) using Tukey's honestly significant difference mean separation ($P \leq 0.05$) with the GLIMMIX procedure. Logarithmic transformation was used for mean separation between fruit counts and seed counts. Arcsine transformation was used for mean separation between germination percentage data.

Results and discussion

Combined over 2021 and 2022, the TH cultivar had the highest number of fruit and seeds per plant compared with other cultivars. TH produced 591.9 fruit per plant, while RW had 79.1 fruit per plant, AL1 had 3.63 fruit per plant, and AL2 had 0.63 fruit per plant (Table 1). TH produced 1257 seeds per plant, RW had 167.0 seeds per plant, AL1 had 6.75 seeds per plant, and AL2 had 1.00 seed per plant. The lower propagule production from the RW, AL1, and AL2 cultivars may result in a lower invasive potential for these cultivars since propagule production is a critical component in promoting invasiveness of *R. frangula* (Berg et al. 2016). The disparity in number of fruit and seeds per plant between the different cultivars is partially due to differences in the duration of fruiting. The TH cultivar was observed to produce fruit from July to November in both years, with most of the fruit harvest occurring in July. In contrast, the narrow-leaved cultivars only produced fruit from August to November.

For both years, plants averaged between 1.51 and 2.12 seeds per fruit for all cultivars (Table 1). The ratio of fruit to seeds did not show any significant trends between cultivars at $P \leq 0.05$. TH did have a significantly greater number of seeds per fruit than both AL1 and AL2 when $P \leq 0.10$. RW also had a significantly greater number of seeds per fruit than AL2 at $P \leq 0.10$. Due to very few fruits of AL2, the accuracy of this difference may be diminished.

For second year fruits, TH had significantly greater seed weight than any other *R. frangula* cultivar tested (Table 1; Fig. 3) with weight of 50 seeds being 1.63 g for TH, 0.95 g for RW, and 1.26 g for AL1. Studies have found for various species that heavier seeds had better survival as compared with lighter seeds of the same species (Cicek and Tilki 2007; Morse and

Table 1. Fruit and seed counts and ratio for four *Rhamnus frangula* cultivars combined for 2021 and 2022, as well as seed weight and germination for 2022.

Cultivar	No. of fruits per plant	No. of seeds per plant	Seeds-to-fruit ratio	Wt per 50 seeds (g)	Seed germination (%) ⁱⁱ
'Columnaris'	591.9 a ⁱ	1257.0 a	2.12 a	1.63 a	47.0 a
'Ron Williams'	79.1 b	167.0 b	2.10 a	0.95 b	0.5 b
Advanced Line 1	3.6 b	6.8 b	1.81 a	1.26 b	3.3 ns
Advanced Line 2	0.6 b	1.0 b	1.51 a	—	—

ⁱ Lower case letters in columns with the same letter are not significantly different by Tukey's honestly significant difference (HSD) test at $P > 0.05$ ($n = 8$; $n = 4$ for the weight per 50 seeds).

ⁱⁱ The seed germination percentage was calculated out of 200 seeds except for Advanced Line 1 where the percentage was calculated out of 30 total seeds which was not significant due to the insufficient number of seeds. No seeds were available to conduct germination tests for Advanced Line 2.

Schmitt 1985; Samreen and Shaukat 2000; Schaal 1980). There is also a correlation of higher seed size of a species in its invasive range than in its native range, suggesting that larger seeds may be favored in the invasive range (Daws et al. 2007). The greater seed size of TH compared with the three narrow-leaved cultivars may

suggest that TH seedlings would have a greater advantage in establishing in unmanaged areas than the narrow-leaved cultivars.

Germination of TH seeds at 47% was greater than germination of RW seeds at 0.51% after 9 weeks (Table 1; Figs. 4 and 5). Comparisons could not be made to AL1 and AL2 for



Fig. 3. Seeds from left to right of *Rhamnus frangula* 'Columnaris', 'Ron Williams', and Advanced Line 1. Ruler tick marks are 1 mm apart.

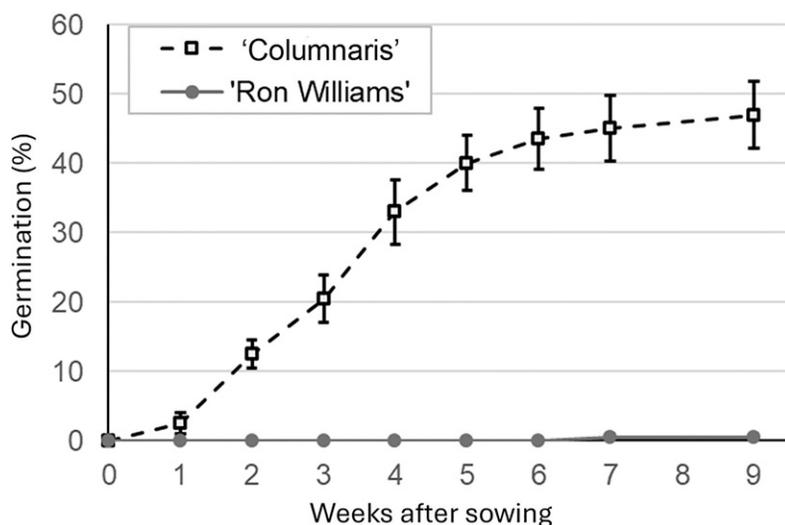


Fig. 4. Germination percent for *Rhamnus frangula* 'Columnaris' and 'Ron Williams' seeds over 9 weeks following sowing.



Fig. 5. Typical germination of 25 *Rhamnus frangula* 'Columnaris' seeds (top) and 25 'Ron Williams' seeds (bottom). The single seedling from 'Ron Williams' seed was the only seedling obtained from that genotype.

germination due to too few seeds produced by both of these cultivars. Thirty ALI seeds were put under the same conditions as seeds from TH and RW, and one of the seeds germinated (3.33%). TH seeds started germinating within 1 week of sowing (Fig. 4). TH continued to germinate throughout the 9 weeks of the experiment, but germination started to slow at about week 6. In contrast, there was only one seed that germinated for RW, which germinated on week 7. The one seed that germinated for ALI germinated in week 2.

The greater and faster germination of TH in comparison with RW suggests a lower invasive potential of RW compared with TH. Germination percentage, as well as the speed of germination, is important in determining invasiveness due to an increased ability to establish in disturbed areas before native plants can establish (Gloria and Pyšek 2016). Furthermore, Gloria and Pyšek (2016) suggest that germination alone is insufficient to determine the invasive potential of plants. Further studies could look at cultivar seed performance at different temperature and light conditions, since *R. frangula* seeds showed variable germination under different temperature and light conditions (Custodio et al. 2023).

Knight et al. (2011) also suggest that a low fecundity of cultivars is insufficient to determine the potential invasiveness of a species due to offspring potentially having similar fecundity to wild-type genotypes. Therefore, fecundity of cultivar offspring could also be considered in future research to be more certain of the noninvasiveness of a cultivar (Knight et al. 2011). We did find that the seedlings of TH and the single seedling of RW had similar morphology in leaf shape, suggesting that the offspring of RW may be more similar to the TH cultivar than the RW parent. It is too small of a sample size to draw any significant conclusions without further study.

Seeds with some scarification, either mechanical, or with a reduced time of sulfuric acid exposure, may also produce different results or higher germination of seeds, similar to the effect of scarification of *R. cathartica* (Kurylo and Endress 2014). *Rhamnus* species, in nature, are dispersed by birds which scarify the seeds naturally.

Conclusions

The results from this study suggest that all three narrow-leaved cultivars may be better options for landscape plants in comparison with TH because of significantly lower fruit and seed production. The lower seed production, lower germination rate, and smaller seed size of RW compared with TH all point to a reduced invasive potential of the RW cultivar. The near zero fruit and seed production of the AL1 and AL2 cultivars over 2 years, and the lower seed weight of AL1 in comparison with TH also suggest a lower invasive potential of these cultivars.

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