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SCIENCE DIRECT®

Flora 201 (2006) 24–31

FLORA

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Seed germination and dormancy in the medicinal woodland herbs *Collinsonia canadensis* L. (Lamiaceae) and *Dioscorea villosa* L. (Dioscoreaceae)

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Received 20 January 2005; accepted 18 April 2005

Abstract

We used a double germination phenology or "move-along" experiment (*sensu* Baskin and Baskin, 2003) to characterize seed dormancy in two medicinal woodland herbs, *Collinsonia canadensis* L. (Lamiaceae) and *Dioscorea villosa* L. (Dioscoreaceae). Imbibed seeds of both species were moved through the following two sequences of simulated thermoperiods: (a) 30/15 °C → 20/10 °C → 15/6 °C → 5 °C → 15/6 °C → 20/10 °C → 30/15 °C, and (b) 5 °C → 15/6 °C → 20/10 °C → 30/15 °C → 20/10 °C → 15/6 °C → 5 °C. In each sequence, seeds of both species germinated to high rates ($>85\%$) at cool temperatures (15/6 and 20/10 °C) only if seeds were previously exposed to cold temperatures (5 °C). Seeds kept at four control thermoperiods (5, 15/6, 20/10, 30/15 °C) for 30 d showed little or no germination. Seeds of both species, therefore, have physiological dormancy that is broken by 12 weeks of cold (5 °C) stratification. Morphological studies indicated that embryos of *C. canadensis* have "investing" embryos at maturity (morphological dormancy absent), whereas embryos of *D. villosa* are undeveloped at maturity (morphological dormancy present). Because warm temperatures are required for embryo growth and cold stratification breaks physiological dormancy, *D. villosa* seeds have non-deep simple morphophysiological dormancy (MPD). Neither species afterripened in a 6-month dry storage treatment. Cold stratification treatments of 4 and 8 weeks alleviated dormancy in both species but *C. canadensis* seeds germinated at slower speeds and lower rates compared to seeds given 12 weeks of cold stratification. In their natural habitat, both species disperse seeds in mid- to late autumn and germinate in the spring after cold winter temperatures alleviate endogenous dormancy.

Keywords: *Dioscorea*; *Collinsonia*; Seed dormancy; Seed germination

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Introduction

Collinsonia canadensis L. (Lamiaceae) and *Dioscorea villosa* L. (Dioscoreaceae), commonly known as stoneroot and wild yam, respectively, are medicinal woodland herbs native to the temperate forests of eastern North America. The rhizomes and roots of *D. villosa* contain diosgenin, a biochemical precursor in the synthetic production of progesterone and other corticosteroids, while the dried rootstocks of *C. canadensis* are used as a diuretic, and the leaves may improve capillary function which aids in the healing of skin wounds (Foster and Duke, 1991). Because of their popularity in Europe and North America as botanical dietary supplements, these

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doi:10.1016/j.flora.2005.04.001

species are wild-harvested from public forestlands (Robbins, 1999), and/or cultivated at small-scales under a forest canopy by private landowners to supply herb companies which manufacture value-added medicinal products from rootstock material (Krochmal, 1968).

With the ever-escalating global demand for botanical forest products (Foster, 1997; Freese, 1998; Vance, 1995), the cultivation of shade-requiring native medicinal herbs under forest canopies is an important conservation management and development strategy in temperate woodlands (Teel and Buck, 2002). Medicinal plant management programs encourage the planting of seed and/or root material to either augment preexisting forest populations, or establish new populations in forests where they are locally absent. Yet reliable information on the population ecology of many botanical forest products is recurrently cited as an impediment to conservation management efforts (Duchesne and Wetzel, 2002; Robbins, 2000; Vance, 2002). Thus, the purpose of this study was to investigate seed germination and dormancy in the increasingly important medicinal forest herbs *D. villosa* and *C. canadensis*.

Although the dormancy-breaking and germination requirements for many polycarpic woodland herbs are well documented (Baskin and Baskin, 2001), little is known concerning the seed germination biology of *C. canadensis* and *D. villosa*. Neither species are cited in reviews on the seed germination ecology of temperate forest herbs (Baskin and Baskin, 1988, 2001). Martin (1946) observed that *C. canadensis* seeds had large, functionally developed embryos, whereas seeds of *D. villosa* had small, functionally underdeveloped, capitate-shaped embryos. When embryos are underdeveloped at maturity, seeds are considered morphologically dormant (MD) because they require some pretreatment for embryos to grow to some critical threshold for full germination (Baskin and Baskin, 2001). Terui and Okagami (1993) reported that *D. villosa* seed air-dried for 50 d germinated at rates <50% at a constant temperature of 25 °C, whereas seeds that were cold stratified for 120 d germinated to >95% over a range of warm constant temperatures (20–25 °C). However, previous studies with *D. villosa* seeds investigated germination behavior under constant laboratory temperatures. Dormancy-breaking and seed germination conditions in nature involve daily temperature oscillations and the natural progression of seasonal temperature changes (Baskin and Baskin, 2001; Probert, 2000; Thompson and Grime, 1983). Consequently, it is essential to evaluate seed germination behavior over a range of simulated thermoperiods and alternating temperature regimes in controlled conditions, and concurrently conduct field experiments to fully characterize dormancy-break (Baskin and Baskin, 2001; Thompson and Grime, 1983).

We experimentally investigated dormancy-break in *D. villosa* and *C. canadensis* by employing a double germination phenology study or "move-along" experiment (Baskin and Baskin, 2003). Based on prior information, we were able to eliminate morphological dormancy (i.e., underdeveloped embryos) as a potential dormancy type in *C. canadensis*. Although fresh seed could be non-dormant (i.e., seed would germinate within 30 d), we hypothesized that *C. canadensis* seeds, which are dispersed in mid-autumn, would have a cold stratification requirement (i.e., physiological dormancy) that would delay germination until favorable spring conditions. Because *D. villosa* embryos are underdeveloped at maturity and appear to require cold stratification for full germination, we hypothesized that seeds would have some form of morphophysiological dormancy (MPD) (i.e., seeds would require some critical temperature for embryo growth and seeds would require some dormancy-breaking pretreatment to germinate). By transferring imbibed seeds through a sequence of thermoperiods that simulate seasonal temperatures in eastern temperate forests, the move-along experimental template would test our hypotheses while also determining the optimum temperatures required for dormancy-break.

Material and methods

Species

Collinsonia is a genus of five species restricted to eastern North America (Gleason and Cronquist, 1991). *C. canadensis* has the broadest distribution within the genus, occurring in early to late-succesional woodlands from Quebec to western Missouri, south to Louisiana and northern Florida, and east to New Hampshire (Gleason and Cronquist, 1991). In early spring, an aerial stem (sometimes two or more) bearing opposite leaves arises from a hard, knotty perennating rhizome (hence the common name stoneroot). Aerial shoots tend to branch near the apex and flowering ramets terminate in divided panicles that bear hermaphroditic flowers from July through September. Flowers are yellow, lemon-scented and obligate xenogamous (Skinner, 1976). Skinner (1976) observed that Bumble Bees (*Bombus* spp.) were the primary pollinators in a naturally occurring population in an Ohio mixed-hardwood forest. The fruit is a four-seeded nutlet that matures during leaf drop in mid-autumn. Seeds are gravity dispersed (Beattie and Culver, 1981).

Dioscorea is a largely pantropical genus with six species occurring in eastern North America, the majority of which are restricted to the southeastern United States. The fleshy tubers and rhizomes of