

Variable seed behavior increases recruitment success of a hardwood tree, *Zelkova serrata*, in spatially heterogeneous forest environments



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ABSTRACT

Variable seed behavior (e.g., seed heteromorphism) plays an important role in enhancing plant fitness by spreading risk in spatially and temporally changing environments. This adaptation is common in herbs but rare in trees. *Zelkova serrata*, one of the most important commercial tree species in Japan, has two types of seeds: shoot seeds that disperse via a seed-bearing shoot and single seeds that disseminate after separating from shoots. To evaluate whether the seeds of *Z. serrata* show heteromorphism, we compared several inherent ecological properties (e.g., seed mass, degree of seed dormancy, and dispersal ability) between the two seed types and assessed the effects of variable seed behavior on recruitment success in forest environments. The degree of seed dormancy did not differ between the two seed types. Seed mass increased with a delayed seed dispersal time but did not differ between the two types at any time. Seed dispersal ability was greater in shoot seeds than in single seeds. These inherent seed behaviors suggest that the two types of seeds do not reflect seed heteromorphism. The results, however, indicate that shoot seeds dispersed larger seeds over much greater distances than did single seeds, particularly at later seed dispersal times, whereas a greater number of single seeds were disseminated near adults. To evaluate the effects of variable seed dispersal ability on seedling establishment in forest environments, we measured seedling performance at different distances (≤ 27.5 m) from adults isolated from one another in the forest understory. Seedling density, survival, and growth did not vary with distance from adults (i.e., there was little negative distance-dependent seedling mortality), which suggests that single seeds can contribute to persistent seedling banks near conspecific adults. To evaluate the advantages of long-distance dispersal of shoot seeds for recruitment on disturbed sites (i.e., gaps), we compared the occurrence and relative abundance of *Z. serrata* among 1100 plots with different slopes; steep slopes are closely associated with gap creation (e.g., landslides). The occurrence and relative abundance of *Z. serrata* increased with slope inclination, which suggests that shoot seeds may enhance recruitment success in disturbed sites along steep slopes, thereby generating *Z. serrata*-dominated forests. This suggests that the variability in seed behavior of *Z. serrata* may enhance recruitment success in spatially heterogeneous forest environments. Knowledge of the unique reproductive behaviors and the wide range of habitats suitable for this species is important for forest managers when making sustainable timber production plans for *Z. serrata*.

1. Introduction

Most plant species exhibit continuous intraindividual variation in seed mass and/or seed morphology (Harper, 1977; Fenner and Thompson, 2005). Different types of seeds (sometimes with accessory parts) can exhibit different behaviors (e.g., dispersal ability, degree of dormancy) as well as so-called seed heteromorphism (Venable et al., 1995; Imbert, 2002). In Asteraceae, central achenes typically achieve greater dispersal than peripheral achenes, although central achenes tend to have a lower degree of seed dormancy (e.g., McEvoy, 1984;

Venable and Levin, 1985; see the review in Imbert, 2002). Variable dispersal distances can enhance plant fitness, particularly in spatially heterogeneous environments, while the disadvantages of short dispersal distances in other morphs are often compensated for by longer periods of seed germination (i.e., spreading offspring over time), reductions in sibling competition, and/or the ability to cope with unpredictably harsh environments (Cheplick, 1996; Venable, 2007). Theoretical models also suggest that variable seed behavior within individuals may enhance fitness, particularly in spatially and temporally unpredictable environments, by evenly spreading the risk for sibling mortality (bet hedging;

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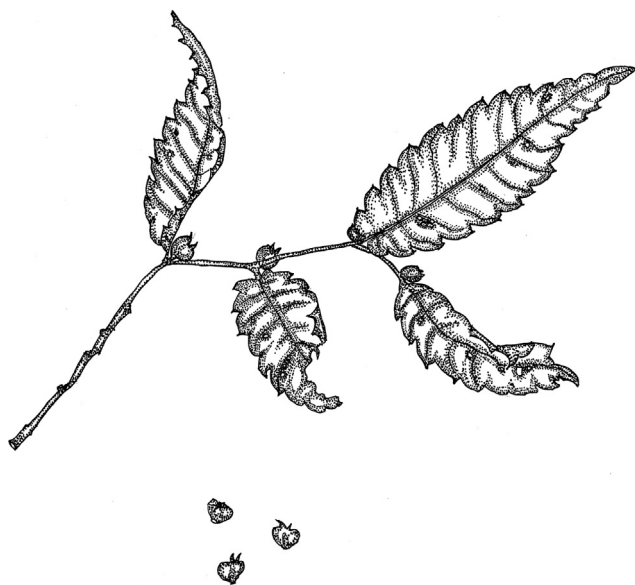


Fig. 1. Two types of seeds in the hardwood tree *Z. serrata*: shoot seeds dispersing with a seed-bearing current-year shoot (a) and single seeds disseminated as single seeds separated from a seed-bearing shoot (b).

Venable and Brown, 1988; Childs et al., 2010). The role of seed heteromorphism in plant fitness has been studied extensively, particularly for herbaceous annuals and monocarpic perennials but rarely for polycarpic plants (Imbert, 2002), likely because polycarpy allows the spreading of reproductive risk over several years (Harper, 1977; Venable and Brown, 1988). In long-lived woody perennials, however, heteromorphic seed behavior can serve as a risk-spreading strategy in changing environments (Zammit and Westby, 1987; Seiwa, 1997; Koyama, 2002; Chiba and Koyama, 2012). Accordingly, the causes and consequences of seed heteromorphism in tree species warrant a more detailed examination.

Zelkova serrata, a deciduous broadleaf tree, has two types of seeds with distinctly different dispersal behaviors (Fig. 1), although the morphological characteristics of the individual seeds (i.e., fruits) do not differ. In this study, we use the term *seed* in its loose sense to mean a single fertilized ovule and the associated structure, including the pericarp. Some seeds are dispersed via seed-bearing, current-year shoots (shoot seeds), whereas others are disseminated as single seeds that separate from the shoots (single seeds; Hoshino, 1990). However, the extent to which the two types of seeds exhibit differences in their inherent ecological properties (e.g., dispersal ability, dispersal timing, seed dormancy, seed germination, and seed size) remains unexplored. If their ecological traits differ, variable seed behavior may enhance fitness in forest environments, as these systems can change substantially through space and time.

Zelkova serrata inhabits temperate forests in northern Japan, and the biotic environments of these ecosystems are spatially heterogeneous. For example, natural enemies such as pathogens and herbivores are more likely to damage and kill plant juveniles that are either growing close to conspecific adults or growing at high densities (i.e., so-called Janzen–Connell [J-C] effects; Janzen, 1970; Connell, 1971). J-C effects have been observed for many hardwood species in temperate forests in Japan (e.g., Tomita et al., 2002; Seiwa et al., 2008; Yamazaki et al., 2009; Konno et al., 2011; Bayandala et al., 2016), temperate forests of North America (e.g., Bennett et al., 2017), and tropical forests (e.g., Harms et al., 2000; Mangan et al., 2010; Comita et al., 2014; LaManna et al., 2017). Recent studies, however, have demonstrated that the strength of J-C effects (i.e., the strength of negative density/distance-dependent seedling mortality) varies substantially among species within forest communities, ranging from negligible to remarkably

strong in both tropical and temperate forests (Comita et al., 2010; Mangan et al., 2010; Johnson et al., 2012; Bagchi et al., 2014; Bennett et al., 2017). If J-C effects are strong, enhanced seed dispersal abilities may increase seedling establishment, as seedlings may be able to escape natural enemies near adults (Janzen, 1970). If shoot seeds of *Z. serrata* show greater dispersal ability compared to single seeds, we can evaluate the extent to which shoot seeds enhance seedling establishment by escaping natural enemies near adults.

In temperate forests, abiotic environments (those that include light, soil moisture, and nutrients) also vary along the gap–understorey continuum with changing topographic conditions (Seiwa, 1998; Nagamatsu et al., 2002; Tateno and Takeda, 2010). Furthermore, gap creation is unpredictable over time (Sakai et al., 1998; Masaki, 1999). *Z. serrata* is distributed across a wide range of habitats with different slopes and is highly dominant in forests located on steep slopes (Shimada, 1994; Nagamatsu and Miura, 1997; Oyama and Seiwa, unpublished data). By contrast, large individuals are less dominant in old-growth forests with gentle slopes (e.g., Terabaru et al., 2004; Hirabuki et al., 2007; K. Seiwa, unpublished data). This, together with the fact that steep slopes frequently experience large disturbances such as landslides (Guariguata, 1990; Seiwa et al., 2013), suggests that *Z. serrata* may populate a wide range of habitats created by different levels of disturbance along a broad range of topographic conditions (i.e., steep to gentle slopes). If so, shoot seeds (which may have greater dispersal ability) may play an important role in recruitment into disturbed sites (i.e., large gaps) along steep slopes. However, empirical data for rare and long-distance dispersal events are limited because of the inherent difficulty of sampling (Nathan and Muller-Landau, 2000). In this study, we evaluated the potential ability of shoot seeds to enhance recruitment success into disturbed sites by investigating the occurrence and relative abundance of *Z. serrata* in 1100 plots that differed in slope inclination.

Zelkova serrata is one of the most important commercial trees in Japan. Most of the large trees that naturally regenerate are harvested, mainly as a result of selective logging of large individuals, which results in a lack of commercial timber in natural forests. Therefore, it is important to investigate reproductive behaviors and habitat selection to ensure more continuous timber production.

In this study, we investigated differences in ecological traits (e.g., dispersal distance, degree of seed dormancy, proportion of filled seeds, and seed mass) between the two seed types (i.e., shoot seeds vs. single seeds) of *Z. serrata*. We also explored the effects of variable seed behavior on recruitment success in spatially changing environments (i.e., habitat choice) in a temperate forest in Japan. We addressed the following questions: (i) To what extent do shoot seeds differ from single seeds in terms of inherent ecological traits? If dispersal ability differs between the two types of seeds, (ii) do shoot seeds enhance seedling establishment success by escaping density/distance-dependent seedling mortality near adults? (iii) Do single seeds enhance seedling establishment near adults via persistent seed or seedling banks? (iv) To what extent does relative abundance increase with increased slope inclination? We also discuss methods of sustainably managing *Z. serrata* to facilitate continuous natural regeneration and continuous timber production.

2. Materials and methods

2.1. Species

Zelkova serrata is a deciduous broadleaf tree commonly found in temperate forests in Japan. This species exhibits polygamonoecious dichogamy; large fruit crops appear every 3–4 years. Within individual current-year shoots, male flowers are observed in proximal positions, whereas hermaphrodite and female flowers are observed in distal positions. Although the male, female, and hermaphrodite flowers are closely proximate in space within individual shoots, anther dehiscence usually occurs later, after the emergence of stigmas (K. Seiwa, personal

observation). Shoots have dedicated abscission zones for the purpose of producing dispersal units of shoot seeds (K. Seiwa, personal observation). Shoot seeds indicates smaller wing loading than single seeds, because terminal velocity was markedly slower for shoot seeds than for single seeds (Data in Brief). There is an evidence that under laboratory conditions the percentage of seed germination is not affected by either the R:FR ratio or seed type (Data in Brief). Similarly, the rates of both outcrossing (approximately 00%) and biparental inbreeding (approximately 0%) did not differ between seed types (Data in Brief).

2.2. Dispersal distance, dispersal timing, seed mass, proportion of filled seeds, and seed dormancy

To examine differences between the two types of seeds (i.e., shoot seeds vs. single seeds) in terms of dispersal distance, dispersal timing, seed mass, and proportion of filled seeds (i.e. seed with a healthy embryo), we installed seed traps (0.5 m²) at a height of 1 m in three directions randomly and at four distances (5, 10, 20, and 30 m) from each of three replicate adults of *Z. serrata*: an adult of diameter at breast height (DBH) 95 cm (Parent 1) and two adults with three trunks each (Parent 2: DBH = 39, 31, and 24 cm; Parent 3: DBH = 63, 5, and 4 cm). In the third adult, seed traps were only set in a single direction because the tree stood at a cliff edge. The three focal adults were open-growing trees, and all were located in meadows or roadsides in the Field Science Center (FSC) of Tohoku University (38°44' N, 140°45' E, alt. 190–200 m) in northeastern Japan. Seed traps were set on 7 September 2011, and seeds were collected at 1- to 3-day intervals until 25 November 2011. At each sampling, the number of seeds in each trap was counted after the seeds were sorted by type (single and shoot seeds). Temporal changes in seed traits (i.e., empty/filled, mass, dormancy status) were assessed after seeds were pooled at 15- to 17-day intervals (first interval: 7–23 September, second interval: 26 September–13 October, third interval: 15–29 October, fourth interval: 2–16 November), although each interval was not contiguous. For each interval, we determined the proportion of filled seeds by cutting open the seeds; the seed dry mass of filled seeds of 100 randomly selected seeds of each type for each adult was measured after seeds were dried to constant weight at 60 °C for 96 h. To examine the degree of seed dormancy, we sowed each type of seed collected from each of the three adults at the four different dispersal times (see above) in a nursery on 6 December 2011. In the nursery, mean monthly temperatures ranged from –1.0 °C (January) to 22.6 °C (August). Mean annual temperature and rainfall were 10.3 °C and 1661 mm, respectively.

In addition, seeds used in the experiment were collected from five seed traps that were randomly placed in a *Z. serrata*-dominated forest (total tree density = 2362 ha⁻¹, total BA = 52.33 m², relative BA of *Z. serrata* = 91.0%, other trees = *Acer palmatum* var. *matsumurae* and *Acer mono*) along a steep slope located 2 km north of the three focal adults. At each dispersal time, seeds were sown after the seeds collected from the three adults and the *Z. serrata* forests were pooled ($n = 13$ –50 per tree or per forest). In both 2012 and 2013, the number of emerged seedlings was monitored weekly from 10 May until late July.

We used a generalized linear mixed model (GLMM) in the `glmmML` package of R to test whether the number of filled seeds (m⁻² day⁻¹) differed by seed type (single vs. shoot seeds), distance from adults (four distance intervals), or dispersal time (four dispersal periods), assuming a Poisson distribution. GLMM analyses were also conducted to test for the effects of seed type and dispersal time on seed mass and percentage of filled seeds, assuming normal and binomial distributions, respectively, as well as the percentage of seed germination, assuming a binomial distribution. Log-transformed data for the observation period were set as an offset variable.

2.3. Seedling establishment at different distances from adults

The seedling establishment of *Z. serrata* was measured in an old-

growth forest on a gentle southeast slope (< 5°; elevation: 180 m) of Mt. Jikyo (peak: 312 m; 38°89'N, 141°00'E) in northeastern Japan (Hirabuki et al., 2007). Trees in the area have been protected from human activity for at least 200–300 years for religious reasons. There were 52 woody species per hectare. The forest is dominated by *Fagus crenata* (relative basal area: 19.6%), *Fagus japonica* (12.6%), and *Carpinus tschonoskii* (12.2%), followed by *Acer palmatum* var. *japonica* (7.2%), *Betula grossa* (5.3%), *Kalopanax pictus* (5.3%), *Cryptomeria japonica* (4.7%), and *Z. serrata* (4.3%; Hirabuki et al., 2007). Annual precipitation is 1212 mm year⁻¹, and mean temperatures in the warmest and coldest months are 24.0 °C and –0.5 °C, respectively, measured at the nearest meteorological station (Ichinoseki, located 12.3 km south-east of the plot, alt. 32 m; Japan Meteorological Agency). Within a 5 ha area of the forest, we selected four reproductive *Z. serrata* trees (DBH: 47.8–60.2 cm) that were isolated from one another. For these four adults, old seedlings (age ≥ 1 year old) of *Z. serrata* were tagged ($n = 1265$), and heights were measured within the circular sector of a central angle of 10° within 30 m of one side centered on the reproductive trees during the period from 22 June to 13 July 2012. Survivorship and vertical growth were measured on 24 July 2013. An exception was made for one adult, for which vertical growth was measured within half of the circular sector (central angle 5°) between 5 and 30 m from the adult, although all old seedlings were measured in the circular sector within 5 m of the adult. All current-year seedlings of *Z. serrata* were tagged ($n = 804$) on 7–9 June 2012, and survivorship was measured on 21 May 2013 in three quadrats of different areas at each assigned distance from the adults (0.25 m² [0.5 × 0.5 m] at both 0–5 and 5–10 m, 1 m² [1 × 1 m] at both 15–20 and 25–30 m) within the circular sector of a central angle of 10° centered on the four reproductive trees. The light conditions (i.e., total direct and diffuse transmittances; mol m⁻² d⁻¹) did not differ by distance from adults (Data in Brief).

The effects of distance from adults on the density and survival of current-year seedlings and the height of old seedlings (age ≥ 1 year old) were analyzed using GLMM. For old seedlings, the effects of distance from adults and seedling height on density, survival, and vertical growth rate were analyzed using GLMM. Seed type, distance, and dispersal time were fixed effects, and adult locality was set as a random effect. The log-transformed quadrat area was set as offset in the analyses of density for both current-year and old seedlings, which assumed Poisson and binomial distributions, respectively. Height and vertical growth rates of old seedlings were analyzed using a normal distribution.

2.4. Occurrence and abundance at different slopes

The occurrence and relative abundance of *Z. serrata* were compared among slopes with different inclinations using a large forest inventory data set (Miyagi Prefecture, 1986). Circular plots of 0.1 ha ($n = 1100$) were randomly established in privately owned deciduous broadleaf forests at lower altitudes (96.4% of the plots were below 600 m) in the Miyagi Prefecture (37°46'N–39°00'N, 140°16'E–141°40'E) of northern Japan from 1981 to 1985. The circular plots were randomly set within the area of 132,400 ha, and the approximate distances between closest nearest neighbor plots and the largest distance between plots were 0.2 km and 5 km, respectively. In this region, most forests on gentle slopes had been previously utilized as fuel-wood coppices or hay meadows but had been abandoned around the 1950 s; they have since regenerated naturally as secondary forests (Seiwa et al., 2012). Thus, the current landscape pattern of the forests is a mosaic of patches of different post-disturbance ages. In each circular plot, two concentric circles (0.04 and 0.02 ha) were established from 1981 to 1985. DBH and height were measured for all woody plants of DBH ≥ 36 cm, DBH ≥ 18 cm, and DBH ≥ 4 cm in the 0.1, 0.04, and 0.02 ha circular plots, respectively. The degree of slope was classified at 5° intervals. Stand age was estimated to be the ages of the largest two or three trees using increment cores, assuming that the trees had become established at a

time when natural or artificial disturbance had occurred. The stems were cored near the surface of the ground. Data from all 1100 plots and from the 131 plots including individuals of *Z. serrata* were used to analyze the occurrence and relative abundance of *Z. serrata*, respectively. Occurrence was defined as 0 or 1. When we observed at least one individual of *Z. serrata* in the plot, occurrence was 1; otherwise occurrence was 0. Relative abundance was defined as the proportion of basal area of *Z. serrata* out of the total basal area of all tree species observed in each plot. Basal area was calculated using all data from the 0.1, 0.04, and 0.02 ha circular plots after the plot area was weighted. Moreover, to evaluate whether the relative abundance of *Z. serrata* changed with time after disturbance, we conducted GLM analyses to test for the effects of stand age and slope inclination on the occurrence and relative abundance (relative basal area) of *Z. serrata*. Occurrence and relative abundance were analyzed using normal and binomial distributions, respectively. All statistical analyses were performed using R version 2.15.2 (R Development Core Team, 2012).

3. Result

3.1. Dispersal distances of filled seeds

The number of trapped single seeds was highest at 5 m and decreased with increasing distance from the adults throughout the dispersal period, although the number of seeds decreased most over time at 5 and 10 m (Fig. 2a; Table 1). By contrast, the number of trapped shoot seeds did not differ across distances (5–30 m), although the number generally increased over time (Fig. 2b; Table 1), resulting in significant interactions between seed type and distance, seed type and dispersal time, and distance and dispersal time (Table 1). The total number of seeds trapped was noticeably higher for single seeds compared to shoot seeds, particularly at 5 and 10 m; however, the difference decreased with increasing distance from the adults (Fig. 2c).

Table 1

Results of GLMM analysis of the effects of seed type, distance, and dispersal time on the number of filled seeds in a hardwood species, *Zelkova serrata*. GLMM: generalised linear mixed model (GLMM).

	Estimate	Z value	SE
Seed type	13.938	114.40 ^{***}	0.122
Distance × time	-0.534	-61.29 ^{***}	0.009
Time	-0.039	-4.05 ^{***}	0.010
Seed type × distance	-0.060	-5.19 ^{***}	0.012
Seed type × time	-1.159	-99.19 ^{***}	0.012
Distance × time	0.043	52.88 ^{***}	0.001
Seed type × distance × time	-0.003	-2.50 [*]	0.001

* $p < 0.05$.

*** $p < 0.001$.

3.2. Seed mass, percentage of filled seeds, and percentage of seed germination

Seed mass, percentage of filled seeds, and seed germination in the nursery increased over time for both single seeds and shoot seeds; minimal differences were observed between the two seed types throughout the dispersal period (Fig. 2d–f; Table 2). Seed germination in the nursery was complete in the spring of 2012, and no seed germination was observed in 2013.

3.3. Seedling establishment at different distances from adults

Both density and percentage survival of current-year seedlings were greatest at 0–10 m from adults and decreased with increasing distance from adults (Fig. 3a and b; Table 3). However, density and survival of old seedlings (age ≥ 1 year old) did not differ by distance (Fig. 3c and e; Table 3), and mean height was highest (20–30 m) when seedlings were farthest from adults (Fig. 3d; Table 3). The vertical growth rate of old seedlings did not differ by distance, although seedling height affected growth rate to some extent (Fig. 3f; Table 3).

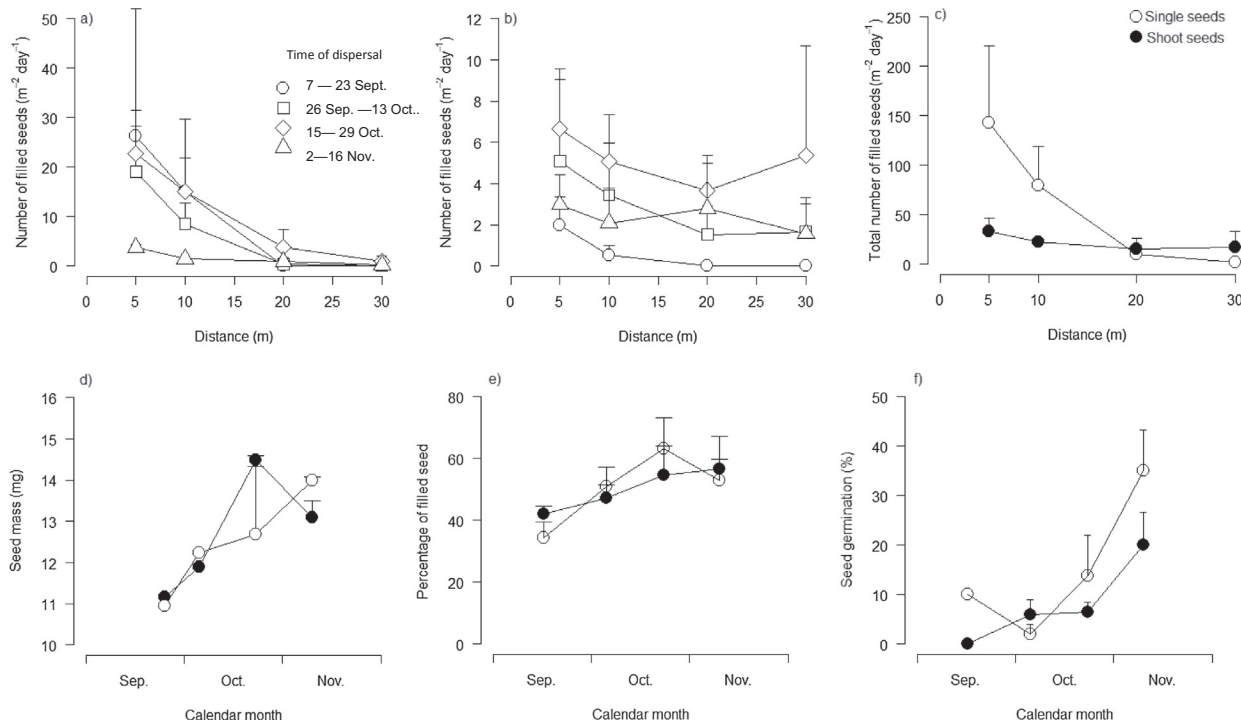


Fig. 2. Number of filled seeds trapped at different distances during four periods of seed dispersal (first period: 7–23 September, second period: 26 September–13 October, third period: 15–29 October, fourth period: 2–16 November) for single seeds (a) and shoot seeds (b). Total number of filled seeds at different distances for single seeds and shoot seeds (c). Change over time in seed mass (d), percentage of filled seeds (e), and percentage of seed germination in the nursery (f) during the four periods of seed dispersal for single seeds and shoot seeds in *Z. serrata*.

Table 2

Results of GLMM analysis of the effects of seed type (single- vs. shoot-seeds), dispersal time and their interaction (dispersal time \times seed type) on seed mass, percentage of filled seeds, and percentage of seed germination in *Zercova serrata*.

		Estimate	Z value ^b (T value ^a)	SE
Seed mass ^a	Seed type	15.672	1.409	11.123
	Time	2.569	4.069**	0.631
	Seed	-1.418	-1.364	1.040
	type \times time			
Percentage of filled seeds ^b	Seed type	-0.117	-0.085	1.383
	Time	0.274	2.641**	0.104
	Seed	0.026	0.196	0.131
	type \times time			
Seed germination ^b	Seed type	2.888	0.547	5.279
	Time	1.582	4.217***	0.375
	Seed	-0.235	-0.484	0.4845
	type \times time			

** P < 0.01.

*** P < 0.001.

3.4. Occurrence and abundance of *Z. serrata* along slope inclinations

Both the occurrence (%) and relative abundance (%) of *Z. serrata* were usually lowest on gentle slopes (< 20°) but increased with inclination, with the highest values occurring on the steepest slopes (> 40°; Fig. 4a and b; Table 4). The occurrence and relative abundance of *Z. serrata* were greater in plots on steep slopes ($\geq 30^\circ$) compared to those on gentle slopes ($\leq 30^\circ$) for all age classes from young (1–30 years old) to old (> 60 years old; Fig. 4c and d; Table 4). Occurrence was lowest at young stages and usually increased with increasing age on all slopes, whereas relative abundance did not differ by stand age (Fig. 4c and d; Table 4).

4. Discussion

4.1. Differences in inherent seed properties between the two seed types

Our results showed few differences between the two seed types (i.e., shoot seeds vs. single seeds) with regard to their inherent ecological properties (i.e., seed mass, percentage of filled seeds, percentage germination, seed dormancy) for the long-lived hardwood tree *Z. serrata*. The seed mass, percentage of filled seeds, and percentage of seed germination were lowest at early dispersal times and increased over time. This strongly suggests that the abscission of seeds from shoots may simply be delayed as a result of reduced seed maturity. This strongly demonstrates that these two seed types do not reflect seed heteromorphism.

However, the number of shoot seeds increased with the time of seed dispersal (Fig. 2b). This, together with the fact that shoot seeds were dispersed over longer distances compared to single seeds, particularly at later dispersal times, suggests that individual shoots dispersed larger seeds far distances from adults. By contrast, a substantial number of single seeds were disseminated near the adults at early dispersal times, although the seeds were smaller in size and lower in germination rate compared to shoot seeds. Our field observations further suggest that this variable seed behavior enhanced the recruitment success of seedlings in spatially heterogeneous environments within temperate forests in northern Japan.

4.2. The role of shoot seeds in recruitment success via long-distance dispersal

In *Z. serrata*, a substantial number of shoot seeds were dispersed at least 30 m from adults, although the dispersal kernels of these seeds

were truncated past 30 m. Fat-tail dispersal kernels strongly imply the potential for much longer dispersal distances in shoot seeds (Clark et al., 1999). Although seed mass increased over time, two or three seeds were shed as single seeds from individual shoots early during the period of seed dispersal, although the seeds were outcrossed, which resulted in a marked decrease in wing loading (i.e., seed mass per shoot area; Augspurger, 1986). This suggests that *Z. serrata* adults may shed outcrossed seeds sparingly early during seed dispersal to increase the dispersal distance of seeds attached to shoots.

Our analysis of the large forest inventory data set further suggests that the greater dispersal ability of shoot seeds enhances recruitment success in disturbed sites. We found that both the occurrence and relative abundance of *Z. serrata* were much greater on steep slopes. This, together with the fact that large disturbances such as landslides occur more frequently on steep slopes compared to gentle slopes (Guariguata, 1990; Nagamatsu et al., 2002; Seiwa et al., 2013), clearly demonstrates that shoot seeds may have a potential advantage for recruitment in disturbed sites on steep slopes via long-distance dispersal. Moreover, *Z. serrata* occurred at large relative abundances (ca. 15–20%) on steep slopes throughout stand development from early to late stages (at least 60 years) after disturbances. This suggests that shoot seeds serve an important role in creating stands dominated by *Z. serrata*, particularly on steep slopes. However, long-distance dispersal cannot be verified from our observations of relatively short dispersal distances (< 30 m; Nathan and Muller-Landau, 2000). More sophisticated methods (e.g., genetic estimates) are needed to verify the successful recruitment of shoot seeds via long-distance dispersal.

A seed-sowing experiment in different topographic conditions revealed that seedling establishment of *Z. serrata* was higher on steep slopes compared to valleys or ridges with gentle slopes, whereas the reverse was true for six other hardwood species (i.e., *Quercus crispula*, *Q. serrata*, *Acer mono*, *Carpinus laxifolia*, *Alnus japonica*, and *A. hirsuta*; Nagamatsu et al., 2002). Because *Z. serrata* elongates its roots horizontally and the root tips are deeply inserted into the soil on steep slopes (K. Seiwa, personal observation), *Z. serrata* could be protected from physical damage by severe surface material flow on steep slopes.

Shoot seeds are also larger in size and have higher germination rates compared to single seeds later on during seed dispersal, which suggests that shoot seeds of *Z. serrata* play an important role in enhancing seedling establishment on steep slopes, on which most other hardwood species can rarely establish.

4.3. The role of single seeds in seedling establishment in the forest understory

Early in the period of seed dissemination of *Z. serrata*, a high proportion of the dispersed seeds were empty, likely because of selective abortion (Owens, 1995; Naito et al., 2005). Simultaneously, a substantial number of filled seeds were also disseminated as single seeds beneath adults. The total number of filled seeds disseminated beneath adults (at distances of either 5 or 10 m from adults) was approximately threefold greater for single seeds compared to shoot seeds. Although single seeds were to some extent smaller and had lower germination rates compared to shoot seeds, all of the single seeds were derived from outcrossing (as were the shoot seeds), although dispersal distance was limited. This suggests that single seeds have a high potential to enhance seedling recruitment near adults.

In the shaded forest understory of the old-growth study forest, minimal differences were observed in the survival and growth rates of *Z. serrata* seedlings across all distances from adults throughout the juvenile stages. Consequently, only small differences were observed in seedling density and vertical height across distances, although current-year seedlings exhibited higher density and survival beneath adults. Although vertical height was also greatest at both 20 and 25 m from adults, large seedlings (1 m < height < 2 m) were only observed in one out of four replicate adults, generating large variance, which

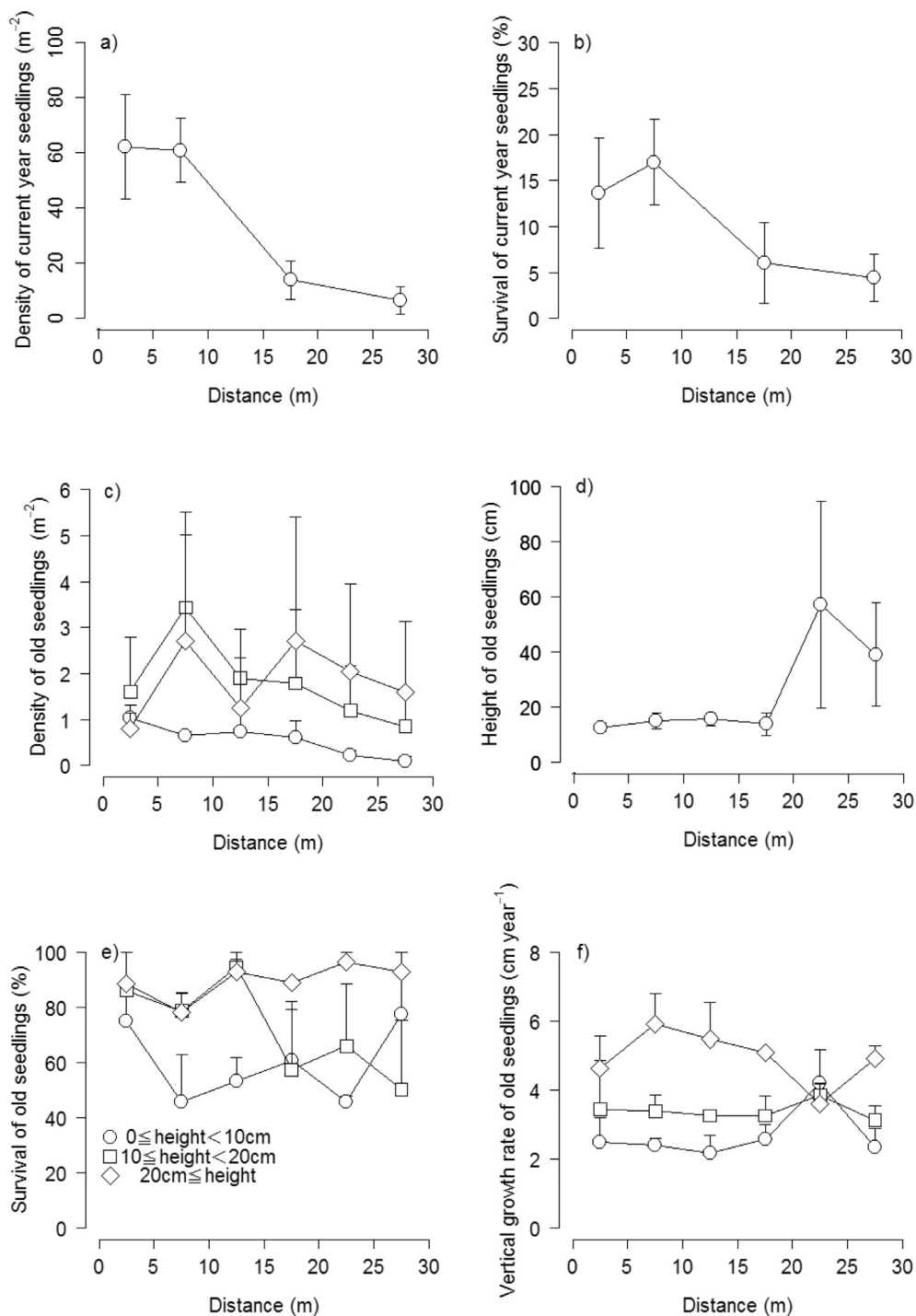


Fig. 3. Density (a) and percent survival (b) of current-year seedlings. Density (c) and height (d) of old seedlings (age ≥ 1 year old). Percent survival and annual vertical growth of old seedlings (age ≥ 1 year old) at different distances for three different height classes (height < 10 cm, $10 \text{ cm} \leq \text{height} < 20$ cm, $10 \text{ cm} \leq \text{height} < 20$ cm).

suggests that vertical height was context dependent. These results imply relatively weak J-C effects (negative distance-dependent seedling survival) in *Z. serrata*. Previous studies have observed weak J-C effects for tree species of relatively high abundance within forest communities (Comita et al., 2010; Mangan et al., 2010; Bagchi et al., 2014). Although the relative abundance of *Z. serrata* was low on gentle slopes, weak J-C effects may have been the result of lower abundances of natural enemies (e.g., pathogens, herbivores; Yamazaki et al., 2009) and/or amortizing negative effects of pathogens via mycorrhizae (Bayandala et al., 2016), even near the adults.

All single seeds germinated during the spring following seed

dissemination, and seed germination even occurred under low light conditions (low R:FR ratios) or in darkness (similar to shoot seeds), which suggests that single seeds may not create persistent seed banks but rather seedling banks near adults (Xia et al., 2016), as mass seeding usually occurs at 1- to 4-year intervals in *Z. serrata* (Yoshino, 2003). As a result, the chance of recruitment to the canopy may be enhanced by single seeds even near adults, although the probability of gap creation may be lower on gentle slopes compared to steep slopes. This strongly demonstrates that single seeds also serve an important role in seedling establishment, particularly in shaded forest understories.

In conclusion, *Z. serrata* inhabits a wide range of habitats from

Table 3

Results of GLMM analysis of the effects of distance on density and survival of current-yr seedlings and height of old seedlings, and the effects of distance, height and their interaction (distance × height) on density, survival and vertical growth rate of old seedlings in *Zelkova serrata*.

		Estimate	Z value	SE
<i>Current-yr seedlings</i>				
Density	Distance	-0.099	-21.07**	0.005
Survival	Distance	-0.047	-3.047**	0.015
<i>Old seedlings</i>				
Density	Distance	-0.007	-1.196	0.006
	Height	0.583	3.571***	0.163
	Distance × height	-0.046	-5.541***	0.008
Height	Distance	0.467	6.16**	0.076
Survival	Distance	-0.0262	-1.062	0.025
	Height	0.037	1.484	0.025
	Distance × height	0	0.38	0.001
Vertical growth rate	Distance	0.075	3.468*	0.021
	Height	0.132	6.58**	0.02
	Distance × height	-0.004	-4.543**	0.009

* P < 0.05.
 ** P < 0.01.
 *** P < 0.001.

Table 4

Results of GLM analysis of the effects of stand age, slope inclination and their interaction (stand age × slope inclination) on occurrence and relative abundance (relative basal area) of a hardwood species, *Zelkova serrata*.

		Estimate	Z-value	SE
Occurrence	Stand age	0.198	3.790***	0.052
	Slope inclination	0.235	3.090**	0.076
	Slope inclination × stand age	-0.015	-1.878	0.008
Relative abundance	Stand age	0.011	1.569	0.007
	Slope inclination	0.032	2.899**	0.011
	Slope inclination × stand age	-0.001	-1.114	0.001

** P < 0.01.
 *** P < 0.001.

gentle to steep slopes in forest environments. This wide distribution is promoted by the existence of two types of seeds with different dispersal abilities. Small numbers of large seeds with a seed-bearing shoot (i.e., shoot seeds) disperse far from adults, facilitating recruitment to steep slopes, whereas small and abundant seeds (single seeds) are disseminated beneath adults, enhancing seedling establishment in a wide range of slope inclinations even in gentle slopes that only rarely

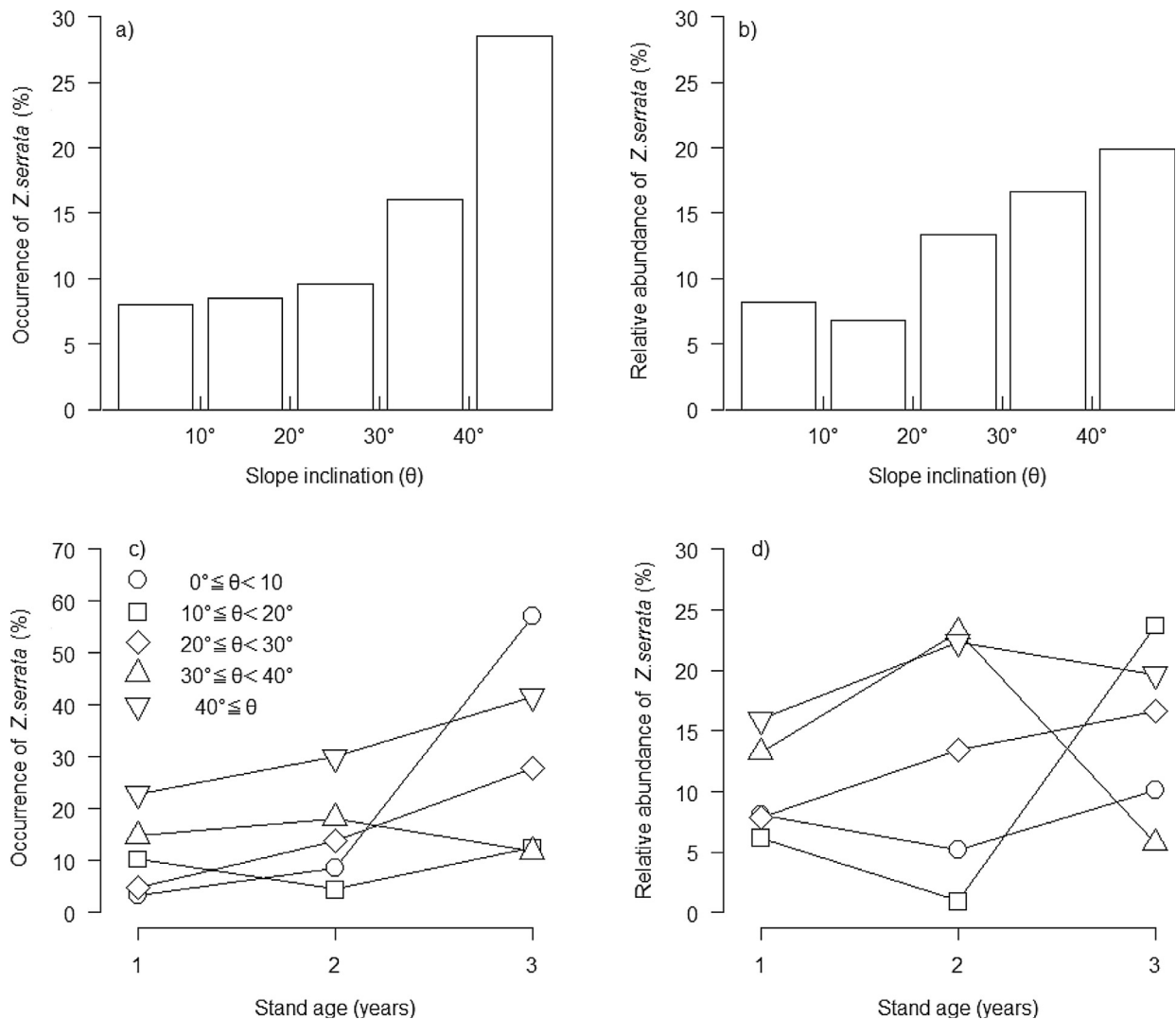


Fig. 4. Relative frequency of the occurrence (a) and relative abundance (b) of *Z. serrata* along a slope inclination of forest stands. Changes with stand age (1: age ≤30 years, 2: 30 years < age ≤60 years, 3: 60 years < age) in the occurrence (c) and relative abundance (d) of *Z. serrata* on different slopes. The occurrence of *Z. serrata* was defined as the percentage of stands that contained at least one individual of *Z. serrata* out of the total number of stands observed (n = 1100). The relative abundance of *Z. serrata* was defined as the proportion of the basal area of *Z. serrata* out of the total basal area of the stands (n = 120).

experience disturbances. The variable dispersal ability of these outcrossing seeds may be advantageous, even for a perennial woody plant such as *Z. serrata*, for establishing in a wide range of habitats within spatially heterogeneous forest environments.

4.4. Implications for management

Because *Z. serrata* is one of the most important commercial trees in Japan, knowledge of the regeneration behavior and subsequent habitat selection of this species is critical for forest managers who wish to develop sustainable timber production plans. *Z. serrata* regenerated successfully in a wide range of topographic conditions from gentle to steep slopes, but the regeneration process and subsequent relative abundance varied greatly according to the slope inclination. According to these results, management methods should be altered. On gentle slopes, a substantial number of the seedlings were successfully established, irrespective of the distance from the adults in the shaded forest understory, and were able to generate persistent seedling banks. Thus, to facilitate seedling recruitment into the canopy stratum, selective cutting would be effective, particularly for canopy trees within a 30 m distance of *Z. serrata* adults. When the slope inclination increased, both the occurrence and relative abundance of *Z. serrata* increased. Although *Z. serrata* frequently dominated on the steepest slopes ($> 40^\circ$), these environments should not be sites for tree harvesting, or even selective cutting, to prevent soil erosion or landslides. On more gentle slopes ($\leq 40^\circ$), selective cutting would be able to produce timber sustainably. However, little is known about the extent to which the abundance of seedlings in the understory changes with slope inclination. Further studies are needed on seedling establishment in a wide range of forest environments and slope inclinations.

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.foreco.2018.02.004>.

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