FOREST FRAGMENTATION EFFECTS ON SEED DISPERSAL, SEED FATE, AND FRUIT PRODUCTION OF DUCKEODENDRON CESTROIDES AND BOCAGEOPSIS MULTIFLORA IN THE BRAZILIAN AMAZON

A Dissertation
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by
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To my father

Dr. Roger E. Cramer

whose advise and wisdom about science, teaching, and graduate school have helped to keep me centered and motivated
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ABSTRACT

In tropical forests, forest fragmentation that alters species interactions between plants and animals, especially mutualisms, will affect the regeneration of trees. Fragmentation reduced *Duckeodendron cestroides* seed dispersal quantity and quality. The percent and distance of dispersed seeds were twice as great in continuous forest (31%, 4.9 m) as in fragments (15%, 2.5 m). Differences were even more exaggerated for each tree’s furthest dispersed seeds. Distributions of dispersed seeds across distance showed more seeds at all distances in continuous forest than in fragments. Dispersal differences were strongest in years when fruit production was high and resulted in a greater number of first-year seedlings at distances far from the parent tree in continuous forests.

Fruit production and post-dispersal seed fate of *Duckeodendron* were also different between fragments and continuous forest. Fruit production, measured by fruit fall, was reduced in fragments, more dramatically in years when fruit production was high. In continuous forest, seeds protected from mammal seed predators or secondary dispersers had high rates of seedling establishment (55%), but the majority of unprotected seeds were rapidly removed (77%). Seed predation was 17 times greater in continuous forest (53%) than in fragments (3%). More seeds in fragments had delayed germination than in continuous forest, resulting in seedlings half as tall as their continuous forest counterparts. Despite these differences in the processes leading up to seedling establishment, there was no difference between the percent of seedlings that established in fragments or continuous forest.

Although fragmentation had dramatic effects on *D. cestroides*, responses of species interactions to fragmentation can vary. Although the number, percentage, distance, and distributions of dispersed seeds were reduced in fragments for the large-seeded *Duckeodendron,*
they were not for the small-seeded *Bocageopsis multiflora*. Seed dispersal of large-seeded species may be more susceptible to fragmentation than small-seeded species because large-seeded species rely on fewer, extinction prone dispersers. Three lines of evidence from other tropical fragmentation research support this hypothesis.

A rapid survey failed to detect differences in fruit production between fragments and continuous forest, probably because small sample sizes and high intraspecific variability caused a Type II error.
CHAPTER 1

INTRODUCTION
Today some of the most rapid deforestation is in the Brazilian Amazon, the world’s largest remaining region of continuous forest (Skole & Tucker 1993, Soares-Filho et al. 2006, Whitmore 1997). Inherent in deforestation is forest fragmentation, the isolation of remaining patches of forest that become surrounded by disturbed or modified habitats. Such dramatic changes to forest size and the surrounding environment must alter the population sizes and interactions of species remaining within the fragment.

Immediately after isolation, forest fragments suffer from reduced species richness of plants because small fragment sizes only allow for a limited representation of species from a highly diverse biota (Laurance et al. 2002). Over time, physical changes such as increased litter accumulation, increased tree mortality from physiological and wind stress (Laurance et al. 1997), and microclimate changes such as decreased humidity and large temperature fluctuations, affect forest fragments along their edges (Laurance et al. 2002). Physical changes visibly and quickly degrade forest remnants along edges and can cause fragments to collapse inward and become even smaller (Gascon et al. 2000). These forest fragments may be too small to be sustainable over time and many animal species will leave the fragments as a result of home range and energy requirements (Gilbert & Setz 2001, Laurance et al. 2002, Rylands 1987, Stouffer & Bierregaard 1995). The degree of isolation of fragments from neighboring fragments, continuous, or secondary forest also limits the movement of animals among fragments (Laurance et al. 2002), further decreasing species richness. The physical effects that cause the reduction in fragment size and the subsequent disappearance of animal species can indirectly affect plant species because many of the disappearing animals can be crucial to processes like pollination and seed dispersal.
Biological effects, such as changes in species composition and abundance, can penetrate 200 to 300 m into the heart of forest fragments (Laurance et al. 2002). Disrupted species interactions, such as seed predation and seed dispersal, may lead to a breakdown of the relationships that sustain forest communities, degrading fragments from the inside and causing them to implode. Although changes to the physical structure and climate of a fragment are more easily recognizable, they may not be as severe as some large-scale biological changes that penetrate deeply into fragments and sever the pervasive species interactions and mutualisms crucial to sustaining tropical communities.

Ecologists are only beginning to understand how fragmentation affects species interactions in tropical forests. Many studies reveal a breakdown between species interactions as a result of forest fragmentation. Fewer insectivorous birds in fragments may be a result of a decrease in their leaf litter arthropod food supply (Stratford & Stouffer 1999). Mutualisms, which are abundant in tropical forests, may be especially at risk as a result of forest fragmentation. Visits by euglossine bees decline in forest fragments (Powell & Powell 1987) resulting in increased self-pollination or decreased fruit set. Although known to persist in forest fragments, populations of ant-plant mutualists are lower in fragments and could be in jeopardy with further disturbance (Bruna et al. 2005). Reduced populations of primates in small fragments leads to smaller populations of dung beetles which then increases rodent seed predation, decreases seedling germination, reduces decomposition, and possibly increases rates of parasite infestation on seeds (Andresen 2003, Andresen & Levey 2004, Klein 1989). In other cases, there is no evidence that fragmentation disrupts relationships between species without compensation by other species. Pollination of Dinizia excelsa actually increased in fragments because introduced Africanized honeybees replaced the original pollinators (Dick et al. 2003).
Ultimately, changes in species relationships, even if compensated for by other organisms, could alter species’ life-history patterns.

In my dissertation I explore the effects of forest fragmentation on several processes; seed dispersal, fruit production, seed predation, germination, and seedling establishment; that influence regeneration of tropical trees. Research was conducted at the Biological Dynamics of Forest Fragments Project (BDFFP), a collaborative research program between Brazil’s Institute for Amazonian Research (INPA) and the Smithsonian Tropical Research Institute (STRI). The reserves of the BDFFP are located on non-flooded upland (*terra firme*) tropical moist forest, about 80 km north of Manaus in the State of Amazonas, Brazil (2°30’S, 60°00’W).

The BDFFP is an ideal place to study fragmentation effects for three reasons. First, the BDFFP is the longest running and most comprehensive fragment project in the world. Between 1980 and 1984, most reserves were isolated from continuous forest by a combination of clear-cutting often followed by burning. Cleared areas were subsequently grazed by cattle and burned again or abandoned to regenerating secondary forest. Today, fragments are maintained by periodically clearing all secondary vegetation from a 100 m border around them. Second, the BDFFP contains fragments of various sizes and nearby continuous control plots for comparison. There are 11 isolated reserves, five 1-ha, four 10-ha, and two 100-ha, spread over three different farms. Lastly, the BDFFP has over 66-ha of mapped forest with at least one 1-ha sample in all isolated reserves. Within these plots, trees > 10 cm DBH are identified, mapped and measured. The database of these trees allows for the easy identification and location of individuals of target species among the 40,000 tagged living individuals.

The first half of this dissertation (Chapters 2 and 3) examines the effects of fragmentation on species interactions and regeneration of *Duckeodendron cestroides*. *Duckeodendron* is an
emergent tree, endemic to the Central Amazon. Like many tropical species, *D. cestroides* is rare, with recorded adult densities of 1.3/ha (Oliveira 1997) and 0.5/ha (BDFFP database). In the BDFFP reserves, *D. cestroides* is documented to be the largest and second longest lived tree species (Laurance *et al.* 2004). Because *D. cestroides* is an endemic, threatened regeneration in fragments can lead to extirpation, and would also result in global extinction.

In Chapter 2, I examine the effects of forest fragmentation on *D. cestroides*’ seed dispersal. *Duckeodendron* fruits are large (5.9 cm long by 3.4 cm wide) pyriform drupes dispersed by primates (records for howler monkeys and bearded sakis), and terrestrial mammals, (records for agoutis and the Guianan squirrel). Over three consecutive years I used wedge-shaped transects to monitor the distance seeds were dispersed away from the parent crown of trees in both fragments and continuous forest. As many primate species at the BDFFP are threatened by fragmentation, I hypothesized that fragmentation would reduce the quantity and quality (distance) of *D. cestroides* seed dispersal. Furthermore, I explored if these trends would remain consistent over several years. Finally, I predicted that reduced seed dispersal in fragments would impact distributions of first-year seedlings.

In Chapter 3, I explore fragmentation effects on other processes important to regeneration of *D. cestroides*: fruit production, seed predation, germination, seedling establishment, and seedling mortality of *D. cestroides* over three years of fruit crops. Fruit production was assessed for trees in fragments and continuous forest by counting fruits surrounding each tree. Post-dispersal processes of seed predation and seedling establishment were examined with 1) an experiment in continuous forest using manipulated seed densities and distances from parent trees that were protected and exposed to mammal seed predators and 2) observations on distributions of naturally dispersed seeds over time.
In Chapter 4, I compare the effects of fragmentation on seed dispersal of a large-seeded species, *D. cestroides*, to that of a small-seeded species, *Bocageopsis multiflora*. Although large animals like the primates that disperse *D. cestroides* are sensitive to fragmentation and often disappear from fragments, smaller animals, like bats and small birds, have been shown to persist in fragments. Animal-mediated seed dispersal of large-seeded species, like *D. cestroides*, is more threatened by fragmentation because there are fewer animals that can disperse and handle their large seeds. In contrast, small seeded species, like *B. multiflora*, which has small fruits (about 0.8 cm diameter); have multiple dispersers, many of which are not sensitive to fragmentation. Given the differences in seed dispersers and their sensitivity to fragmentation, I predicted that there would be differences in the effects of fragmentation on seed dispersal of the two species. In addition, I construct a hypothesis with three lines of evidence from dispersal and fragmentation studies worldwide, supporting the generalization that fragmentation affects large-seeded species, like *D. cestroides*, more than small seeded species, like *B. multiflora*.

In my final chapter (Chapter 5) I investigate the effectiveness of a rapid survey in examining the effect of fragmentation on fruit production. Fruit production is dependent on pollination and resource availability, so reduced fruit production can indicate species that are the most threatened by fragmentation. Rapid surveys that can effectively assess populations in fragments and continuous forest would be helpful in identifying threatened species from species rich tropical forests.
CHAPTER 2

FOREST FRAGMENTATION REDUCES SEED DISPERSAL OF DUCKEODENDRON CESTROIDES, A CENTRAL AMAZON ENDEMIC

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7
Introduction

Fragmentation of once continuous forest can have dramatic effects. Animal and plant populations often decline and become less resilient to disturbance, with many species becoming extirpated from fragments (Ferraz et al. 2003, Laurance et al. 2002, Silva & Tabarelli 2000). Additionally, some tree species in fragments exhibit increased mortality (Laurance et al. 2000) and decreased recruitment (Benitez-Malvido 1998).

The effects of fragmentation may be exacerbated in tropical forests where biotic interactions prevail, interconnecting many species, especially through mutualisms (Gilbert 1980). Reduced plant regeneration in tropical fragments may be related to declines in animal species that are vectors for pollination and seed dispersal (Benitez-Malvido 1998, Howe 1977, Howe 1984b, Tabarelli et al. 2004). Fragmentation can also cause changes in frugivore behavior, resulting in reduced seed dispersal (McConkey & Drake 2006). How forest fragmentation alters mutualistic processes, such as seed dispersal, is a priority for research (Laurance et al. 2002), given the paucity of studies (Bruna et al. 2005, Cordeiro & Howe 2003, Dick et al. 2003, Galetti et al. 2006, Wright & Duber 2001).

Here, I present evidence for changes in the seed dispersal of a rare tropical tree, *Duckeodendron cestroides* Kuhlm. (Solanaceae), that are caused by forest fragmentation. *Duckeodendron* fruits are consumed by primates and terrestrial mammals such as peccaries, agoutis, and pacas. However, many of these medium to large vertebrates are rare in or absent from forest fragments in the Amazon Basin (Gilbert & Setz 2001, Peres 2001, Timo 2003). For *D. cestroides*, I hypothesized that the decline in seed dispersers would result in reduced seed dispersal and seedling recruitment. Specifically, I asked the following: (1) Does forest fragmentation result in the reduced quantity (number of seeds) and/or quality (seed distance),
sensu Schupp (1993), of seeds dispersed away from parent \( D. cestroides \)? (2) Are fragmentation effects on seed dispersal constant from year to year? (3) Are first-year seedling distributions a reflection of seed dispersal distributions or are they affected by distance-dependent mortality, sensu the Janzen-Connell hypothesis?

**Methods**

**Study Site**

This study was conducted at the Biological Dynamics of Forest Fragments Project (BDFFP), a collaborative research program between Brazil’s National Institute for Amazonian Research (INPA) and the Smithsonian Tropical Research Institute (STRI). The BDFFP reserves are located on non-flooded upland (terra firme) tropical moist forest, about 80 km north of Manaus in the state of Amazonas, Brazil (2°30’S, 60°W) (Lovejoy & Bierregaard 1990). Mean annual rainfall is 2,651 mm, with a distinct, but mild, dry season (Laurance 2001). Mean annual temperature is 26° C (Bruna 2002). Density of trees > 10 cm DBH averages 613 stems/ha and trees > 60 cm DBH are uncommon with only 11 stems/ha (Laurance 2001). Alpha diversity is high with over 1000 species of trees in 61 families and 288 genera (Gascon & Bierregaard 2001).

Between 1980 and 1984, 11 fragments (five 1-ha, four 10-ha, and two 100-ha) were isolated from continuous rainforest by clear-cutting, often followed by burning around each fragment, although the specific histories of each fragment are unique (Gascon & Bierregaard 2001). The BDFFP monitors tree species in these reserves every six to thirteen years.

**Study Species**

*Duckeodendron cestroides* (Khulm.) is an emergent tree, endemic to the Central Amazon. With adult densities of 0.5/ha (BDFFP database) to 1.3/ha (Oliveira 1997), *Duckeodendron* exhibits a rarity representative of nearly 80% (404/513) of the tree species in the Central
Amazon (Oliveira 1997). Since its description in 1925, the genus *Duckeodendron* has been placed into various families, including its own, the Duckeodendraceae (Kuhlmann 1947). More recently, molecular data have assigned this species to a monospecific genus in the Solanaceae (Fay *et al.* 1998). To date, there have been no published studies on the ecology of *D. cestroides*.

Fruiting of *D. cestroides* peaks in the dry season (Oliveira 1997) although ripe fruits can be found on the forest floor throughout the year and old fruits without pulp can persist on the ground for months. Fruits are pyriform drupes approximately 59 mm long by 34 mm wide (J. M. Cramer, unpublished data). When ripe, the fruit possesses a thin, orange exocarp (the pulp) that covers a thick, fibrous mesocarp of hairs resembling the husk of a coconut in texture (Santiago-Valentin & Olmstead 2003). The pulp has a strong, stinky odor. Fruits with pulp are twice as heavy as those without pulp (37.5 g and 18.6 g, respectively, J. M. Cramer, unpublished data.). Beneath the mesocarp’s fibrous hairs, the seed is further protected by a very hard, stony endocarp. Most fruits contain one seed (Kuhlmann 1947) although 2.3% of sampled fruits (*N* = 478) produced two stems indicating either two seeds per fruit or two sprouts from a single seed (J. M. Cramer, unpublished data).

Although *D. cestroides* fruit is anecdotally considered poisonous (C. F. da Silva, pers. comm.) or dispersed by extinct megafauna (Ribeiro *et al.* 1999), fruits are regularly removed from the forest floor (J. M. Cramer, unpublished data). The fruit’s size, odor, bright color and the well protected seed suggest that medium to large mammals are the primary seed dispersers (Roosmalen 1985, Van der Pijl 1982). *Duckeodendron* fruits at BDFFP have been recorded in the diets of two primate species—red howler monkeys (*Alouatta seniculus*) (Neves & Rylands 1991, P. Stouffer pers. obs. 2003) and bearded sakis (*Chiropotes sagulatus*) (S. Boyle, pers. obs. 2006) — and are probably also consumed by black spider monkeys (*Ateles paniscus*) and brown
capuchins (*Cebus apella*). Arboreal mammals may strip the pulp from the fruit while in the canopy and then drop the fruits to the ground where they may be further handled and dispersed by terrestrial mammals. It has been suggested that acouchies (*Myoprocta* spp.) are probable dispersers (Ribeiro *et al.* 1999), and I believe terrestrial mammals contribute to secondary dispersal or to seed predation, as 73% of experimentally placed seeds were removed within 7 weeks (Ch. 3). Numerous seeds at the study site have tooth marks on the seed endocarp and lack seed kernels.

**Field Data Collection**

In the 2002 dry season (July-October) I located 11 *D. cestroides* with at least 30 fruits on the forest floor: four trees in 10-ha fragments, two in 100-ha fragments, and five in continuous forest (tracts >1,000,000-ha). Trees were selected based on the size of their 2002 fruit crop and location relative to other fruiting conspecifics. In 2003 another continuous forest tree was added to the census and in 2004 a nearby treefall forced us to exclude a previously sampled *Duckeodendron* from a 100-ha fragment. The BDFFP’s phytodemographic database allowed us to accurately determine the location of fruiting as well as nonfruiting conspecifics within the census plots.

Two transects established around each individual in July and August, 2002, were used to estimate seed dispersal for each tree (Figure 2.1). I used wedge-shaped transects to accommodate increased dispersal area and decreased seed density as distance from the parent tree increased, similar to seed trapping methods used by Clark *et al.* (2005). Wedge-shaped transects eliminate census bias based on distance-dependent seed density. I chose this method because past seed dispersal studies have shown conventional seed trap methods failed to catch a single seed from as many as 88% of species over a 10-year period (Hubbell *et al.* 1999) and that
FIGURE 2.1 Transect layout for seed dispersal census, illustrated for Tree#5 from continuous forest, to scale except transect length which is abbreviated to conserve space.
in a given year, only 7% of species have seeds that reach at least 10% of randomly placed seed traps (Muller-Landau 2001).

Transects were placed to accommodate irregular fruiting patterns throughout the crown and to avoid overlap with other fruiting *D. cestroides*. I employed four criteria in the following order of priority: 1) avoid transects of other focal trees, 2) capture the highest density of fruit on the ground under each tree, 3) avoid fragment edges and trails, and 4) maximize the distance between transects of a given focal tree without violating the first three criteria. These criteria were established to maximize the quantity of fruit censused without transect overlap or other anomalies that would cause changes in disperser behavior (forest edges) or the likelihood of seed disturbance (researcher trails). When two fruiting individuals were within 50-100 meters of each other, both were used only if sampling transects could be placed in directions radiating away from each other to minimize transect overlap.

Each census year, transects originating at the crown center, regardless of trunk location, were extended until no seeds were found in three to four consecutive distance classes. Therefore, the length of each transect was independent of the lengths of all other transects for the same or other focal trees and for different years. Terminating transects according to seed availability allows censusing for dispersed seeds without bias towards seed density. Transects of equal lengths may be biased because greater effort is given to searching for rarer occurrences of seeds around trees where seed dispersal is reduced. The crown edge was delineated at three points on each transect, at both edges and the transect median. Because tree crowns are naturally variable in shape, it was not unusual for each of these three points to be a different distance away from the transect origin (Figure 2.1). Distance from the crown was marked independently along both transect edges and the transect median in the following intervals: 1-m intervals from the
crown edge to 10 m from the crown, 2-m intervals from 10-40 m from the crown, and 3-m intervals beyond 40 m from the crown. Each wedge-shaped transect has a 45˚ base angle, so together the two transects constitute 25% of the tree’s potential dispersal area for distances censused. Both transects were pooled for each tree for all analyses.

During the dry seasons of 2002, 2003, and 2004, I searched the leaf litter within transects to locate and record the positions of seeds relative to the parent’s crown. Seed locations were individually marked with a small, wooden stake and subsequently checked 1-2 months later (2002 and 2003, only). Disappearance of seeds after the initial census could represent either secondary dispersal or seed predation. All first-year seedlings, determined by the color and quality of the seed’s mesocarp, were marked and counted concurrently with each year’s census. These seedlings provide a snapshot of seed germination rates as virtually all *Duckeodendron* seeds that do not suffer from predation germinate (Ch. 3).

Given the low density of *Duckeodendron* at the BDFFP, the 10 and 100-ha fragments were tested for differences in the percent of seeds dispersed. These fragment sizes were found to be similar, so they were combined into a single class, “fragments”, for comparison to trees in unfragmented, continuous forest.

**Data Analyses**

I used several analyses to test for differences in seed dispersal between forest fragments and continuous forest. Primarily, I asked what proportion of seeds (dispersal quantity) were dispersed at least 1 m beyond the crown, what was the mean distance (dispersal quality) of those dispersed seeds, and what was the shape of the dispersal curve, always looking for variation by forest type (fragments versus continuous forest) and by year. I also focused on the most successful dispersal each year by repeating the mean distance analyses for the five furthest
dispersed seeds. Finally, I examined the cumulative effect of fragmentation on dispersal quality by asking how many seeds were dispersed more than 10 m beyond the crown over all years. Because the same individual trees were repeatedly measured every year, the variable “tree nested in forest type” was included as a random effect in the model. In SAS (PROC MIXED and GLIMMIX) random effects define repeated measures with constant variance and covariance. Details of the statistical treatments follow.

A Generalized Linear Mixed Model (GLMM, SAS, PROC GLIMMIX) analysis (SAS Institute Inc. 2005) with a negative binomial distribution and natural log transformation was used to test for differences in the number of seeds dispersed more than 1 m past the crown of parent trees for fragmentation and year effects. Negative binomials are appropriate for natural count data because populations are often associated with overdispersion as a result of correlated errors in time and space. A negative binomial distribution accounts for overdispersion by allowing for larger variances relative to the mean, whereas Poisson distributions assume variances and means are equal. The total number of seeds found was included in this analysis as an offset variable to adjust for different fruit fall recorded for each tree. An offset variable is the same as a covariate with a fixed coefficient of one.

Two-factor ANOVAs (SAS, PROC MIXED) were used to test for differences in the total number of seeds found in the combined transects of each tree and the mean distance from the crown of dispersed seeds by forest type and year. The dependent variables, total number of seeds (+1) and mean distance from the crown of dispersed seeds were natural log transformed to achieve normality and equal variances.

A GLMM analysis with a Poisson distribution and natural log transformation was used to examine the effects of fragmentation and year on the mean dispersal distance of each tree’s five
furthest dispersed seeds in each year. I also examined the cumulative effect of fragmentation on dispersal distances for each tree over all three years on (1) the number of seeds dispersed beyond 10 m from the parent crown ($X^2$ analysis) and (2) the mean distance of the five furthest dispersed seeds for all three years combined (SAS, PROC GLIMMIX).

A Random Coefficient Regressions (RCR) was used to describe the number of seeds found as a function of distance from the crown, assuming a Poisson response (SAS, PROC GLIMMIX). A RCR is a generalized linear mixed model that fits subject-specific models (individual trees) about a population-averaged model (fragments or continuous forest) (Moser 2004). RCR models are similar to analysis of covariance (ANCOVA), but the latter assumes fixed effects for the covariate, whereas an RCR assumes that the regression coefficient for one or more covariates are a random sample from some population of possible coefficients (Littell et al. 1996). I compared the regressions with intercepts that were allowed to vary randomly among trees and years. A one-tailed test determined if forest fragments had reduced dispersal relative to continuous forest. Variable-sized sampling areas, resulting from the irregular crowns, were included in the analysis as an offset variable. The equation of a triangle was used to extrapolate the sample areas of the crown and each distance interval (sum of two triangles). I removed the number of seeds found under the crown of each tree and the zeros at all distances after the last seed was found in each transect to limit the regressions to dispersed seeds only. Removing crown data eliminated large drops in seed numbers between the crown edge and first distance class after the crown (0-1 m). Removing excess zeros at the end of the dispersal curves eliminated variable sized tails and standardized the data for all regressions.

Finally, two-factor ANOVAs (SAS, PROC GLIMMIX) were employed to compare the total number of seedlings and percent of germinated seeds in fragments versus continuous forest
for three different distance classes: under the parent crown, crown edge out to 10 m, and beyond 10 m from the crown to transect end. To achieve the best fit for normality and equal variances, the total number of seedlings was fit with a Poisson distribution and a natural log transformation, whereas the proportion of germinated seeds was fit with a binomial distribution and a logit transformation. The total number of seedlings and the percent of germinated seeds were used as response variables to determine if dispersal distance had an effect on first-year seedlings. When there was a significant interaction between the independent variables, separate effects of each variable were tested.

When appropriate, a Kenward-Rogers adjustment to the degrees of freedom was added to analyses to provide a conservative Type I error control for small sample sizes. The Kenward-Rogers adjustment results in fractional degrees of freedom seen throughout the results. Except for the Chi-squared test, the analyses for this paper were generated using SAS software, (SAS Institute Inc. 2002).

**Results**

**Seed Dispersal**

Over three years I marked and counted a total of 3767 *D. cestroides* seeds in the dispersal transects. Seed totals varied among years – 1805, 734, and 1226, respectively, in 2002, 2003 and 2004. There were 2752 seeds in continuous forest and 1013 in fragments, with 17 tree censuses over all three years for each forest type.

I tested for two potentially confounding factors, distances transects were sampled and total number of fruits sampled from fruit fall, that might influence my interpretation of dispersal differences between continuous forest and fragments. The distance at which transects were searched was not significantly different between forest fragments (mean = 8.0 m ± 1.1) and
continuous forest (mean = 7.8 m ± 1.1; $F_{1,33} = 0.03$, $P = 0.86$). However, for all three years, total number of seeds per tree (fruit fall) was greater in continuous forest ($\ln(\text{seeds}) = 4.7 ± 0.33$) than in forest fragments ($\ln(\text{seeds}) = 3.6 ± 0.33$; $F_{1,10} = 5.75$, $P = 0.04$). Seed totals also fluctuated over the three years ($F_{2,18} = 14.23$, $P = 0.0002$) with the fewest seeds in 2003. However, there was no significant interaction between these two variables, forest type and year ($F_{2,18} = 1.08$, $P = 0.36$). Given the absolute difference in fruits recorded between fragments and continuous forest, subsequent tests that included both dispersed and undispersed seeds were adjusted for fruit fall.

Only 16 percent of seeds were dispersed further than 1 m past the crown edge in forest fragments, compared to 31 percent in continuous forest (Table 2.1). Year was also significant

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Figure 2.2 Continuous forest trees had a significantly greater (A) percent of seeds dispersed more than 1 m past the crown ($N = 34$ trees over three years), (B) dispersal distances of seeds from the parent crown ($N = 31$ trees over 3 years), and (C) dispersal distances of the five furthest seeds/tree/year ($N = 32$ trees over 3 years) than forest fragments. Asterisks above paired bars indicate significant differences between continuous forests and forest fragments for that year. Back-transformed LSMeans (with 95% CI) are presented here, but significance tests were based on transformed means.
with a smaller percent being dispersed in 2003 (13%) than in 2002 (26%) or 2004 (32%) (Table 2.1, Figure 2.2A), and there was no significant interaction between year and forest type.

Seeds were dispersed shorter distances beyond the edge of the crown in fragments (2.5 m) than in continuous forest (4.9 m) and shorter distances in 2003 (2.5 m) than in 2002 or 2004 (4.4 m and 4.0 m, respectively, Table 2.1). A significant interaction between forest type and year (Table 2.1) showed that continuous forest trees had greater mean dispersal distances than forest fragment trees in 2002 and 2004 ($F_{1,15} = 8.66$, $P = 0.01$ and $F_{1,15} = 11.72$, $P = 0.004$, respectively) but not in 2003 ($F_{1,15} = 0.11$, $P = 0.74$) (Figure 2.2B).

For the five furthest dispersed seeds of each tree in each year, differences between fragments (3.0 m) and continuous forest (9.3 m) were even more exaggerated (Table 2.1). Year was again significant with greater dispersal distances in 2002 (8.7 m), and 2004 (6.7 m) than in 2003 (2.7 m; Table 2.1). Again, a significant interaction between forest type and year (Table 2.1) showed that the means were different between continuous forest and fragments in 2002 ($F_{1,11.4} = 10.6$, $P = 0.007$) and in 2004 ($F_{1,13.6} = 7.0$, $P = 0.02$), but not in 2003 ($F_{1,20.4} = 0.67$, $P = 0.42$) (Figure 2.2C).

Over the cumulative three years, the five furthest dispersed seeds averaged 18.7 m beyond the parent crown in continuous forest versus 5.0 m in forest fragments (GLMM $F_{1,10} = 9.44$, $P = 0.01$). In addition, 20 times more seeds were dispersed > 10 m from the parent crown in continuous forest (344 seeds) than in forest fragments (17 seeds) — a difference much greater than expected from the number of seeds recorded (2752 and 1013, respectively) in each forest type ($\chi^2 = 200$, $P < 0.0001$).

The number of seeds at each distance interval followed a negative trend with distance that was non-linear and highly variable among trees and years (Figure 2.3). For trees, seed number
Figure 2.3  The density of dispersed seeds per 10 m$^2$ across distances from the maternal tree in fragments and in continuous forest for years 2002-2004, (Figures 2.3A-C) and the RCR population estimates for all years combined (D). Trees in fragments typically have a lower density of seeds across all distances than trees in continuous forest. Each line represents an individual tree, solid shapes represent trees where seeds were only found under the crown (A-C). Note that the x-axis distance is greatly reduced in 2003 (B). The y-axis for the RCR estimate (D) is reduced because it only accounts for seeds dispersed beyond the crown.
decreased with distance from the crown, but not monotonically as it was common to find many seeds in one distance interval, no or few seeds in the next distance interval, and then another increase in seed number. Despite this variation, the regressions provided significant linear fits for most trees in most years when seed numbers were high, although not when seed production was low, as in 2003 (data not shown).

The RCR analysis showed that the number of seeds dispersed over all years for both continuous forest and forest fragments decreased with increasing distance from the edge of the tree crown ($F_{1,154} = 424, P < 0.0001$). A comparison of fragments versus continuous forest revealed that the seed distributions of continuous forest and fragments differed in their intercepts ($F_{1,10.2} = 4.4, P = 0.03$) but not in their slopes ($F_{1,140} = 0.01, P = 0.46$). As the dispersal regression for continuous forest was above the regression for fragments, the number of dispersed seeds was greater at all distances (Figure 2.3D). Because the regressions were performed on log-transformed distances, the same slopes signify that the decline in seed number with the log of distance in fragments was similar to the decline in continuous forest, even though the fragments started with much fewer seeds at the intercept (crown edge). Finally, the furthest dispersed seeds were much further from the crown in continuous forest than in fragments (Figure 2.3D). This last result is not an artifact of variable transect lengths, because I performed the RCR regressions on transect lengths uniformly truncated to 6 m from crown edge, the maximum distance with seeds for every tree, and obtained the same results.

Seed removal (secondary dispersal or seed predation) between censuses in 2002 and 2003 was a small percent of the total (mean = $4.7\% \pm 2.36$). The mean percent of seeds removed between censuses in 2003 ($7.1\% \pm 4.5$) was more than triple that in 2002 ($2.1\% \pm 0.77$).
However, the total number of seeds removed in 2003 \((N = 66)\) was only double that removed in 2002 \((N = 32)\).

**Seedling Establishment**

Although there was no difference in the number of seedlings per tree found in continuous forest versus forest fragments (Table 2.1), seedling numbers were dependent on distance from parent trees (Table 2.1, Figure 2.4A) with significantly fewer seedlings 0-10 m from the crown than under the crown (Bonferroni pairwise comparisons \(P = 0.05\)). A significant interaction between distance and forest type indicated that seedling numbers declined at different rates for each forest type (Table 2.1). Separate tests by forest type showed significantly more seedlings under the crown and 0-10 m from the edge of the crown than beyond 10 m in continuous forest \((F_{2,18.7} = 6.73, \ P = 0.006)\). In forest fragments there were significantly fewer seedlings 0-10 m from the crown than under the crown \((F_{2,65} = 5.79, \ P = 0.005)\). Continuous forest and forest fragments differed in the number of seedlings only for the 0-10 m distance class \((F_{1,37.7} = 7.85, \ P = 0.008)\). Therefore, the number of seedlings declined immediately after the crown in forest fragments but only beyond 10 m from the crown in continuous forest. The net effect of this differential decline in seedlings with distance was no seedlings beyond 10 m from the crown over three years for fragment trees in contrast to the 22 seedlings from nine different trees in continuous forest.

The proportion of seeds that became seedlings showed a significant interaction between forest type and distance class from parent tree (Table 2.1). Separate tests by forest type showed no distance effect in forest fragments \((F_{2,93} =0.60, \ P = 0.55)\) but significant differences across distance classes in continuous forest \((F_{2,93} =4.13, \ P = 0.02)\) with a higher percent of seedlings germinating beyond the crown than under the crown (Figure 2.4B). Separate tests by distance
Figure 2.4 (A) Number of seedlings and (B) percent of germinated seeds in continuous forest and forest fragments at three distance classes from parent trees. Data show LSMeans ± 95% CI. Asterisks above paired bars indicate significant differences between continuous forest and forest fragments for that distance class.
classes, showed that the percent of germinated seeds was not significantly different between forest types for 0-10 m from the crown ($F_{1,39.4} = 0.03, P = 0.87$) but was nearly so under the crown ($F_{1,28} = 3.69, P = 0.07$) (Figure 2.4b), and obviously different at 10 m+ from the crown, as there were no seedlings this far in fragments.

**Discussion**

**Seed Dispersal**

Over all three years, both the quantity (proportion) and quality (distance) of dispersed seeds in continuous forest were double those in fragments. In addition, the distance of the furthest dispersed seeds in continuous forest was triple that in fragments. Finally, predicted dispersal curves for each forest type show more seeds dispersed to all distances from parent crowns in continuous forest than in forest fragments.

The reduced seed dispersal of *D. cestroides* in forest fragments may be attributed to the declines of terrestrial and arboreal mammalian dispersers for this species. The responses of primates and large mammals to fragmentation at the BDFFP coincide with the changes in dispersal I observed in *Duckeodendron*. Of the six primate species at the BDFFP, black spider monkeys (*Ateles paniscus*) and bearded sakis disappeared from the forest fragments immediately after isolation and remained absent for at least 10 years after isolation (Gilbert & Setz 2001). Bearded sakis are a known consumer of *D. cestroides* fruit. The other confirmed consumer of *Duckeodendron*, red howler monkeys, are less sensitive to fragmentation, occasionally occupying 10-ha fragments (Gilbert & Setz 2001). In a recent study, sand traps (ten, 0.25m² for a week) in fragments and continuous forest at the BDFFP recorded many fewer medium and large terrestrial mammals in fragments (9 records) than in nearby continuous forest (60 records).
Thus, it appears that reduced seed dispersal in forest fragments resulted from overall mammal reductions, not the disappearance of a single species — an anticipated result given the asymmetry of most dispersal mutualisms (Bascompte et al. 2006) and the switching of dispersers among preferred fruit species (Herrera 1998, Levey & Benkman 1999).

Rates of *Duckeodendron* fruit removal 1-2 months after initial censuses indicated that terrestrial animals were important secondary dispersers or seed predators. Over six weeks, a camera trap aimed at ten experimentally placed seeds under the canopy of a *D. cestroides* in a nearby continuous forest site recorded the Guianan squirrel (*Sciurus aestuans*, 6 visits), the brown four-eyed opossum (*Metachirus nudicaudatus*, 5 visits), and a Marmosa or *Marmosops* sp. (1 visit) (Yabe et al. 1998). Over a two-week period, my camera traps at two continuous forest sites photographed a margay (*Leopardis wiedii*) and three grey-winged trumpeters (*Psophia crepitans*) at *Duckeodendron* fruits, and two agoutis (*Dasyprocta sp.*), a capybara (*Hydrochaeris hydrochaeris*), and one Guianan squirrel (*Sciurus aestuans*) in the bait vicinity.

Other potentially important terrestrial mammals, such as acouchies, agouties, pacas and rats (*Proechimys* spp. and *Oryxomys* spp.), are all present in fragments at the BDFFP. These species tend to hoard seeds, so they could be important secondary dispersers, although I did not track secondary dispersal. Scatter-hoarding has been shown to increase seed survival and escape from seed predation by rats (Asquith & Mejia-Chang 2005). Undetected scatter-hoarded seeds in fragments could counter my results by increasing the number of dispersed seeds in fragments and reducing the difference I saw in dispersal between fragments and continuous forest. If so, more scatter-hoarded seeds in fragments should have resulted in more seedlings in fragments, but my results showed more seedlings in continuous forest. Also, fragmentation may reduce populations of scatter-hoarders, resulting in a lower probability of scatter-hoarded seeds in fragments (Galetti
et al. 2003). Therefore, it is unlikely that undetected scatter-hoarded seeds in fragments would change my interpretation of fragmentation effects on seed dispersal.

Fruiting of tropical trees is often highly variable with years of high fruit production interspersed with years of low or no fruit production (Herrera et al. 1998). Here, for *D. cestroides*, fruit fall fluctuated over the three years of my study; total fruit production in 2003 was about half of 2002 and 2004 (40% and 60%, respectively). When fruit production was high, differences between fragments and continuous forest in the proportion of seeds dispersed and dispersal distances were significantly different or nearly so. In these years, seeds in continuous forest were found up to 46 m (2002) and 30 m (2004) from the parent tree, triple and double, respectively, the maximum dispersal distance (14 m) in any fragment. In contrast, in 2003 when fruit production was low, there were no detectable differences between continuous forest and forest fragments in the percent of seeds dispersed or distance of dispersed seeds. Therefore, differences in dispersal between fragments and continuous forest were sharp during years of high fruit production, exactly when dispersal was important.

Reduced dispersal differences in 2003 may be a consequence of reduced *D. cestroides* fruit production. In 2003 a crash in fruit production in neighboring French Guiana resulted in frugivorous animals leaving the area (P. M. Forget, pers. comm.). Although reduced fruit crop sizes often do not affect the distance that seeds are dispersed (Bleher & Böhning-Gaese 2000, Laska & Stiles 1994), they appear to determine the attractiveness of a fruiting tree and, consequently, the likelihood of disperser visitation (Jansen et al. 2004). The lack of dispersal differences between forest types in 2003, when fruit production was low, may reflect poor visitation leading to poor dispersal.
Just as low disperser visitation could explain the lack of dispersal for all trees in 2003, it could be argued that smaller fruit crops of trees in forest fragments could result in poor disperser visitation (Jordano 1995, Saracco et al. 2005) and explain the reduced seed dispersal I recorded here. Forest fragmentation could reduce fruit production, perhaps as a result of poor pollinator visitation and resulting low fertilization. I found that greater fruit fall was recorded for trees in continuous forest than trees in forest fragments over the course of three years. To be conservative in the analyses, I incorporated the total number of seeds counted as an offset variable (SAS Institute Inc. 2005) in the analysis, comparing the proportion of seeds dispersed in continuous forest to forest fragments while adjusting for fruit fall of each tree in each year. A greater proportion of seeds were dispersed in continuous forest than in forest fragments for all three years, indicating that the differences in seed dispersal found here are strong despite modest differences in fruit production. Therefore, if poor disperser visitation is responsible for low dispersal in fragments relative to continuous forest, it is likely a result of changes in the disperser community, not simply differences in recorded fruit fall.

Although I grouped 10 and 100-ha fragments as a result of limited sample sizes, both percent dispersal and the distance where the last seed was found had responses in 100-ha fragments that were intermediate to those in 10-ha fragments and continuous forest in years of high fruit production (2002 and 2004) but not years of low fruit production (2003) (analyses not shown). These trends suggest that shifts in seed dispersal depended on fragment size, and that fragment area should be considered in dispersal studies wherever possible.

A critical assumption in this study is that the seeds found in the dispersal transects were assigned to the appropriate parent tree. A recent seed dispersal study on Simarouba amara in Panama found that the nearest adult was not always the maternal parent to a seedling (Hardesty
et al. 2006). Without molecular markers, I cannot be definite of seed parentage and therefore of dispersal rates and distances for *Duckeodendron*. However, all of the parent trees were isolated from other fruiting conspecifics or were placed with their transects radiating away from fruiting conspecifics. The latter case occurred only once in this study, a pair of trees in continuous forest where any errors in assigning parentage would have underestimated dispersal distances. The net effect of underestimated dispersal distances would have yielded a greater actual difference in dispersal between continuous forest and fragments than I observed.

A limitation of sampling for dispersed seeds on the ground is that rare instances of long distance dispersal are missed. In addition, as I did not monitor fruit removal from tree crowns, I could not account for seeds removed directly from trees and dispersed beyond my transects. Such differences could be problematic if dispersers in fragments behave differently from those in continuous forest (McConkey & Drake 2006). Seeds dispersed far beyond parent crowns and my transects may represent individuals of the highest dispersal quality, if they have the greatest likelihood of survival to adulthood because they escape distance-dependent mortality. I did analyze the tail of the distributions by looking at the five furthest dispersed seeds, and they exhibited more exaggerated differences between fragments and continuous forest than did all seeds.

Reduced seed dispersal resulting from forest fragmentation has been reported in three other studies, all in the tropics. Cordeiro and Howe (2003) found fragmentation reduced dispersal agents and seed removal of the endemic tree *Leptonychia usambarensis* in the East Usambara Mountains of Tanzania, although they did not report direct counts of dispersed seeds. Wright and Duber (2001) used direct counts of seeds to show that dispersal of *Attalea butyraceae* in Panama was less in areas where mammal populations were reduced by both hunting and
fragmentation. Similarly, Galetti et al. (2006) found that in Brazil’s fragmented Atlantic forest the probability of seed removal of the large-seeded endemic palm, *Astrocaryum aculeatissimum*, decreased as mammal dispersers were eliminated by hunting and fragmentation. These studies all linked reductions in dispersal in hunted fragments to reductions in seedlings numbers or densities. Results from this study further support these findings that forest fragmentation does reduce seed dispersal, here based on direct counts and on the distances seeds were dispersed in areas experimentally fragmented, where hunting was not a factor.

**Seedling Establishment**

I estimated the effect of distance-dependent mortality on seeds and seedlings by looking for newly germinated seedlings shortly after fruit fall and comparing the percent of germinated seeds across distance classes. The Janzen-Connell hypothesis states that predators should operate more intensely at distances closer to the parent tree. In forest fragments, the pattern of seedling germination contrasted with the Janzen-Connell hypothesis as the percent of germinated seeds under the crown was not different from the percent at 0-10 m beyond the crown, and there were no seeds in the 10 m+ class (Figure 2.4B). While the percent did not change, there were more absolute numbers of seedlings under the crown because there were more seeds there (Figure 2.4A). In continuous forest, distance-dependent mortality may be more important for *Duckeodendron* as the percent of germinated seeds 0-10 m beyond the crown was greater than under the crown.

Differences in the influence of distance from the parent tree on germination in continuous forest and forest fragments resulted in differences in the absolute numbers of seedlings. Although both forest types showed that the number of seedlings declined with distance, this decline occurred much closer to parent trees in fragments than in continuous forest (Figure
2.4A). The accelerated decline in seedling numbers with distance from parent crowns in fragments resulted in significantly fewer seedlings 0-10 m from the parent crown. This difference in seedling numbers with distance can be attributed to the number of seeds dispersed over all distances: Increased seed dispersal in continuous forest resulted in greater numbers of seedlings at distances beyond the crown. In fragments, the low number of dispersed seeds coupled with their decline in number as distance increased, resulted in few seedlings 0-10 m from parent crowns and a complete absence of seedlings more than 10 m beyond parent crowns. Although I did not detect a change in the percent of seeds that germinated in fragments one to two months after fruit fall, seedlings close to parent trees might still show increased susceptibility to distance-responsive herbivores or pathogens over longer periods. In fragments, where all seedlings are concentrated around parent plants, distance-dependent seedling mortality could have an extreme effect, eliminating nearly all seedlings in fragments.

Conclusions

Fragmentation effects of disrupted mutualisms on trees may be difficult to detect because the results take longer to surface in long-lived species. In addition to reduced seed dispersal and fewer seedlings in fragments shown here, fragment trees may also be affected by decreased probability of fertilization (Cascante et al. 2002), increased seed abortion (Chacoff et al. 2004), increased seed predation by vertebrates (Francisco et al. 2002) and decreased seed predation by insects (Cascante et al. 2002, Janzen 1978). By studying the mutualistic processes involved in plant regeneration, such as seed dispersal and first-year seedling establishment, changes that forecast the next generation can be detected before they appear in the adult community.
CHAPTER 3

FOREST FRAGMENTATION EFFECTS ON THE REGENERATION OF DUCKEODENDRON CESTROIDES, A RARE TROPICAL TREE
Introduction

Regeneration of rare tropical trees in fragmented forests may differ from recruitment in continuous, unfragmented forest. Unfortunately, little is known about the processes contributing to seedling regeneration in fragments, although some species exhibit reduced densities of seedlings in forest remnants (Benitez-Malvido 2001).

Regeneration of trees involves complex ecological interactions, from flower and fruit production to seed dispersal and seedling establishment, all of which may be sensitive to forest fragmentation. In fragments, mutualisms such as pollination and seed dispersal may be disrupted (Aizen & Feinsinger 1994, Cordeiro & Howe 2001, Cunningham 2000a, Powell & Powell 1987, Ch. 2), seed predation can increase (Francisco et al. 2002, Wright & Duber 2001) or decrease (Cunningham 2000b, Janzen 1978), and the physical environment may be altered (Kapos 1989, Laurance et al. 2002). For a few species, fragmentation of tropical forests is known to reduce seed dispersal and recruitment (Cordeiro & Howe 2003, Galetti et al. 2006, Ch. 2, Wright & Duber 2001). However, for most tropical trees, little is known about fragmentation effects on fruit production or post-dispersal seed fate, both important determinants to successful recruitment (Forget 1993).

Fruit production is dependent on successful pollination and available resources. Most tropical trees rely on animal pollinators (Bawa 1990), so disruption of pollination perhaps poses the greatest risk for trees in fragments (Didham et al. 1996, Gilbert 1980, Howe 1977, 1984a, Tabarelli et al. 2004), although exotic pollinators may substitute for natives (Dick 2001). Fragmentation isolates rare tropical trees, many of which are self-incompatible (Bawa 1974, 1979, 1990), thereby decreasing pollination and lowering fruit
set. In addition, changes to the environment within fragments can physically stress trees, causing lower rates of fruit set or greater rates of seed abortion (Chacoff et al. 2004).

Seed predators can strongly influence post-dispersal seed fate. Small terrestrial mammals, many acting as seed predators, have been shown to increase in density and diversity in fragments (Malcom 1988, 1997). Increases in their abundance can increase seed predation, potentially reducing recruitment. In contrast, medium-sized rodents, like agoutis and acouchies, which function as seed predators and dispersal agents, exhibit reduced densities in forest fragments (M. Jorge, pers. comm.). Likewise, insect seed predators, such as bruchid beetles, may be affected by habitat destruction (Chacoff et al. 2004, Janzen 1978). Negative impacts of fragmentation on seed predators might potentially benefit seedling establishment, although predicting the outcome of such complex interactions is difficult.

Seed and seedling fate in fragments may also be impacted by changes in density- and distance-dependent mortality. Reduced seed dispersal in fragments leads to greater densities of seeds closer to parent plants (Cordeiro & Howe 2003, Galetti et al. 2006, Ch. 2, Wright & Duber 2001). If density- and distance-dependent seed predators and pathogens disproportionately increase seed and seedling mortality close to parent plants (Connell 1971, Janzen 1970), then mortality could be exaggerated in fragments. However, almost nothing is known of how fragmentation and distance effects interact to influence the seed and seedling communities (Stevenson 2000).

I monitored fruit production and post-dispersal seed and seedling fate for the large-seeded tree, Duckeodendron cestroides (Kulhm.), a Central Amazon endemic. Fruit production was monitored by counting dispersed seeds in fragments and continuous
forest over three years. Post-dispersal seed fate was monitored in an experiment and with concurrent observations of naturally dispersed seeds. In the experiment, artificially dispersed seeds were used to examine seed removal and seedling establishment of seeds at different distances from adults, at different densities, and unprotected and protected from mammals by exclosures. Naturally dispersed seeds in fragments and continuous forest were monitored in distance classes from nearest adult conspecific to observe rates of 1) seed predation, 2) seedling establishment, 3) seedling survival, and 4) seedling growth. When important, I also examined the effect of time, within and between years of fruit production, on the naturally dispersed seeds. I predicted that fragmentation would alter fruit production, seed predation, and seedling establishment, but given the complexity of ecological interactions, I did not predict the direction of these outcomes.

**Methods**

**Study Site**

All field research was conducted in the reserves of the Biological Dynamics of Forest Fragments Project (BDFFP), a collaborative project between Brazil’s National Institute for Amazonian Research (INPA) and the Smithsonian Tropical Research Institute (STRI). The reserves are located on non-flooded upland (*terra firme*) tropical moist forest about 80 km north of Manaus in the state of Amazonas, Brazil (2°30’S, 60°W). Mean temperature is 26°C (Bruna 2002) and mean annual rainfall is 2,651 mm with a distinct dry season between June and October (Laurance 2001). Alpha and beta tree diversity are high with over 1000 species of trees (Gascon & Bierregaard 2001). Tree density is also high, averaging 613 stems/ha for trees > 10 cm DBH (Laurance 2001).
Between 1980 and 1984, most reserves at the BDFFP were isolated from extremely large tracts of forest (> 1,000,000 km²) by clear-cutting, often followed by burning, although the specific isolation histories of each fragment are unique (Gascon & Bierregaard 2001). Today there are 11 isolated reserves (forest fragments): five 1-ha, four 10-ha, and two 100-ha, spread over three different farms and numerous control plots located within the remaining large tracts of unfragmented forest, hereafter called “continuous forest”. Isolation of the fragments is maintained by periodically clearing a 100-m wide belt of all secondary vegetation around each fragment.

**Study Species**

*Duckeodendron cestroides* (Solanaceae) is an emergent tree endemic to the Central Amazon. Typical of many species in this region, *D. cestroides* is rare with recorded adult densities of 0.5/ha (BDFFP database) to 1.3/ha (Oliveira 1997).

*Duckeodendron* is the largest and second longest lived tree documented in the BDFFP reserves (Laurance et al. 2004).

*Duckeodendron* fruits are pyriform drupes approximately 59 mm long by 34 mm wide. When ripe, the pulp is orange and has strong musty odor. Fruiting peaks during the dry season months of April-July (Oliveira 1997) although new fruits with pulp can be found on the forest floor during other months. As fruits age, the remaining fruit is easily distinguishable by its unique fibrous mesocarp. The seed of each fruit is surrounded by a hard stony endocarp except at the fruit’s apex, which is covered only by the fibrous mesocarp. Seedling shoots and roots emerge from the seed at the apex. After falling to the ground, seeds or their exocarps can persist on the forest floor for months, or even years.
Fruit Production

*D. cestroides*’ fruit production was measured by fruit fall in fragments and continuous forest for three consecutive fruit crops. In July 2002, wedge-shaped transects radiating from the crown center were established around 11 adult *D. cestroides*, six in fragments and five in continuous forest (Ch. 2). In August of 2002, 2003 and 2004, near the end of *D. cestroides*’ annual fruiting, the leaf litter within all transects was searched to locate all seeds from that year’s fruit crop. Data on fruit fall were obtained through these seed counts (Ch. 2). An analysis of covariance was used to determine if fruit fall was affected by either fragment size (10-ha, 100-ha, and continuous forest – the latter assigned a value of 1000-ha) or fruit crop year (2002, 2003, and 2004). Fruit fall and fragment size were log-transformed to linearize the fits.

Experimental Seed Removal And Seedling Establishment

I initiated an experiment to examine the effects of seed density, distance from parent crowns, and mammal predation on *D. cestroides* fruit in continuous forest. At the start of the dry season of 2003 (May-July), three individuals of *D. cestroides* were chosen as focal individuals because they were not currently being censused for seed dispersal or seed fate of naturally dispersed seeds. These three trees were at least 100 m from other fruiting *D. cestroides*, were not on a slope, and had at least 40 fruits that showed no seed predation. All fruit and seeds under and around each tree were collected for use in the experiment and to create a study area where no other *Duckeodendron* seeds were present.

I manipulated seed density, distance from the parent crown, and exposure to mammal predators to examine their effects on seed removal and germination. Around each tree, 14 line transects radiating from the crown center were placed at 26° intervals
(Figure 3.1). Each transect was divided into three distance classes: 1) under the crown, 2) 10 m outside the crown, and 3) 35 m outside the crown. At each distance class, seeds were placed in groups of three different densities: 1, 5 and 10 seeds/0.5 m$^2$. These distances and densities were chosen to represent the minimum, mean, and maximum distributions of naturally dispersed seeds in continuous forest (Ch. 2). In addition, each seed density was either exposed to potential mammal secondary dispersers and predators, or protected with a mammal exclosure. Exclosures were constructed of chicken wire and secured to the ground with metal stakes so that seeds were at least 10 cm from the edge of the cage. Exclosures were 0.5 m tall and came to a peak to prevent leaf litter accumulation. Densities of five and ten seeds were represented by a single replicate per distance, but densities of one seed were replicated five times at each distance (Figure 3.1). One adult had enough seeds for the experiment, but for the other two adults about 50% of experimentally placed seeds were collected from a different parent and evenly distributed throughout the treatments.

Each seed was assigned a unique number and marked with a small wooden stake at its base, for identification at subsequent censuses. Seeds were checked for removal and seedling establishment at five subsequent censuses: first at about 3 weeks (2 – 4 weeks), then at 5 weeks and 7 weeks, and later at 63 and 93 weeks. In all analyses week was considered as a categorical variable, as it represented the censuses, but in the figures, the actual number of weeks is presented to indicate time since the start of the experiment.

I used two-way ANOVAs to evaluate the percent of seeds removed and the percent of seeds established as seedlings as a function of two fixed effects – exclosure presence and week. Residuals were modeled for heterogeneous variances between
Figure 3.1 An example of treatment layout for experiment on manipulated seed distributions. Dots represent groups of seeds that vary according to distance from parent tree, density, and exposure to mammal predators. Location on transect represents three distances from parent crown, under the crown, 10 and 35 m from crown edge. Size of dots represents number of seeds in each treatment; • 1 seed, ○ 5 seeds, and □ 10 seeds. Closed dots represent treatments protected from mammal predators with wire cages and open dots represent unprotected treatments.
treatments with and without exclosures. Results were heavily weighted towards removal when seeds were unprotected and towards seedling establishment when seeds were protected. Therefore, I analyzed effects of density and distance on seedling establishment only for treatments with exclosures, and on removal only for treatments without exclosures, using analyses with fully crossed three-way ANOVAs to test for effects of seed density, distance, and week. The analysis for removed seeds was modeled with heterogeneous variances on distance.

Seeds without exclosures were often quickly removed; so, two different focal trees were given replacement seeds at the 35 m distance at week 5. These replacements were seven replicates of single seeds and two replicates each of 5 and 10 seeds (a total of 37 seeds). Since replacement seeds entered the experiment at week 5, I used a two-way ANOVA to test if these seeds had different rates of removal or seedling establishment over the duration of the experiment, with week included in the analyses. Seed removal was modeled with heterogeneous variances on replacement versus original seeds.

Between observations at weeks 7 and 63, some wire exclosures collapsed, leaving seeds no longer completely protected from mammals. These seeds were included in analyses only through week 7. However, I opportunistically determined the effect of fallen cages versus intact cages on subsequent removal and seedling establishment by a two-way ANOVA with exclosure status (fallen or intact) and week as fixed main effects (PROC MIXED, SAS); the analysis for removed seeds was modeled with heterogeneous variances on exclosure status.

All two-way ANOVAs included random effects of individual seeds nested within the focal tree because the same seeds were measured multiple times. This type of random
effect defines repeated measures with constant variance and covariance in SAS (PROC MIXED). The three-way ANOVA included random effects of the focal tree, the interaction between the focal tree and distance, and individual seed nested within the interaction between the focal tree, distance and density. A Kenward-Rogers adjustment to the degrees of freedom was added to all analyses to provide conservative Type I error estimates for small sample sizes and resulted in fractional degrees of freedom throughout the results. All analyses were generated using SAS software (SAS Institute Inc. 2002).

**Fate of Naturally Dispersed Seeds**

I monitored seeds through time for two major seed fates: predation and seedling establishment. At censuses several months apart, each seed was marked with a small wooden stake to help relocate it in later censuses. However, seeds were not individually identified, so they could have been moved in and out of the transects, away from the stakes, and sometimes stakes were lost.

I tested the influence of four main effects on the fate of naturally dispersed seeds. First, I tested the effect of fragment size by comparing trees from fragments, 10-ha and 100-ha combined as the “fragment” class, to trees from continuous forest. Second, I tested the effect of distance from parent crown for three classes: under the parent crown, 0 to 10 m beyond the crown, and 10 m+ beyond the crown. Third, I tested for temporal variation among three fruit crop years: 2002, 2003, and 2004. Fourth, I tested for temporal variation within each fruit crop in number of months (nested in fruit crop year) following this initial census. These months, which represent time from fruit fall, varied irregularly and somewhat erratically, depending on my ability to census. Also, with three fruit crops across three years, earlier cohorts were monitored longer than later ones.
Rodent seed predation was monitored for the 2003 and 2004 fruit crops, but not the 2002 fruit crop. Rodent predation was identified by a lengthwise gash penetrating the endocarp, tooth marks, and the absence of a seed kernel. This measure of seed predation is different from seed removal in the experiment, described above, where seed removal represented removal by either seed predators or dispersal agents. Here, percent seed predation was found by dividing the number of seeds with predation by the total number of seeds at each census. When there were no seeds found, the data point was excluded from the analysis. Few trees had seeds dispersed into the 10 m+ distance class in fragments and continuous forest for the 2003 and 2004 fruit crops, so I eliminated that distance class from the analysis on percent predation for all years.

Seedlings and their heights were monitored throughout the study for all three fruit crop years, as each seedling was flagged with a unique number.

I defined the variable “percent seedlings” as the number of seedlings in a transect divided by the total number of seeds and seedlings present at that census (times 100). Both the divisor and the dividend varied through time, as seeds could have been removed from or moved into the transects, and as there were new seedlings and deaths of seedlings from prior censuses. When there were no seeds found, the data point was excluded from the analyses.

The “percent of new seedlings” was defined as the number of new seedlings, found for the first time at each census, divided by the total number of seedlings recorded throughout the study for each transect-distance combination. The absence of new seedlings resulted in zero percent new seedlings for a census. However, transect-distance combinations that never had seedlings were excluded from the analysis.
Similarly, the “percent of dead seedlings” was defined as the number of newly dead seedlings at that census divided by the total number of seedlings recorded throughout the study. Because there was only a single seedling in fragments beyond 10 m, I eliminated the 10 m+ distance class for the analyses on percent of new seedlings and percent of dead seedlings.

All four of the main effects were analyzed for percent seed predation, percent seedlings, percent of new seedlings, and percent of dead seedlings, with fully crossed ANOVAs (SAS, PROC MIXED). The same trees were repeatedly measured for each fruit crop and at various distance classes, so random effects incorporating these components were included in the analyses to specify repeated measures. All percentages were arcsine transformed to increase the normality of the data. When there were significant interactions, I used simple main effects tests to look for differences between fragments and continuous forest by distance, month, and fruit crop year or differences across distance classes by fragmentation, month, and fruit crop year.

Time of seedling survival was determined by the number of months between the first and last censuses the seedling was alive. Therefore, my estimates of seedling survival are conservative and can be 0 months when seedlings were not recorded for more than one census. I used fully crossed two-way ANOVAs (PROC MIXED, SAS) to estimate the effects of fragment size and distance from the parent crown on time of seedling survival and on height. These models included random effects of individual parent tree nested within fragmentation type. Number of months was natural log-transformed to achieve normality. The distance class 10 m+ was excluded from the analysis on seedling survival because there were seldom any seedlings in fragments at
this distance. I also ran ANCOVAs to estimate the effects of fragmentation and density on seedling survival and growth. Random effects were similar to the ANOVAs used for percent predation and percent seedling establishment.

Again, Kenward-Rogers adjustments to the degrees of freedom were incorporated in analyses to accommodate small sample sizes and all analyses were generated using SAS software (SAS Institute Inc. 2002).

Results

Fruit Production

In fragments, fruit fall was lower, and varied more among the three years than in continuous forest. The log-log relationship between fragment size and fruit fall was highly significant with less fruit fall in smaller forest fragments \( F_{1,26.7} = 18.44, P = 0.0002 \). Yearly variation was in the intercept \( F_{2,25.1} = 5.73, P = 0.009 \) but not in the slope \( F_{2,25.1} = 1.56, P = 0.23 \). Pairwise comparisons showed that fruit fall was significantly lower in 2003 than in 2002 and 2004 for both 10 and 100-ha fragments (Bonferroni adjustment \( P < 0.01 \)) but there were no differences among years in continuous forest (Bonferroni adjustment \( P > 0.05 \)). Log-log regressions showed significant positive relationships of fragment size on fruit fall for 2002 \( R^2 = 0.39, P = 0.04 \), 2003 \( R^2 = 0.45, P = 0.02 \), and 2004 \( R^2 = 0.40, P = 0.04 \) (Figure 3.2). These results were essentially unchanged, if continuous forest was changed from an arbitrary assignment of 1000-ha to 10,000-ha (2002: \( R^2 = 0.37, P = 0.05 \); 2003: \( R^2 = 0.39, P = 0.04 \); 2004: \( R^2 = 0.38, P = 0.04 \)) or to 100,000-ha (2002: \( R^2 = 0.36, P = 0.05 \); 2003: \( R^2 = 0.36, P = 0.05 \); 2004: \( R^2 = 0.37, P = 0.05 \)).
Figure 3.2 Positive relationship between fruit fall and fragment size showing both predicted curves and true data points of trees for three fruit crop years. Values shown are back-transformed from log-log relationships.

Experimental Seed Removal and Seedling Establishment

When mammals had access to *D. cestroides* seeds, removal was high and increased through time ($F_{4,366} = 3.45, P < 0.01$) (Figure 3.3). After 93 weeks, the percent of unprotected seeds removed (mean=77% ± 5) was 45 times that for protected seeds (mean=1.7% ± 0.9). Nearly 60% of the seeds were removed in the first 3 weeks and 72% after 5 weeks (Figure 3.3). Through week 93, rates of removal were greater for seeds that were present at the start of the study (79% ± 3.1) than for replacement seeds set out at week 5 (30% ± 7.4) ($F_{1,63.7} = 16.07, P = 0.0002$); there was no significant interaction with week ($F_{2,25.4} = 0.70, P = 0.50$).

Where exclosures remained intact, seed removal rates were close to zero (Figure 3.3). However, 18 exclosures, spread across all distance classes, collapsed between weeks 7 and 63. Consequently, 44 exclosure seeds were no longer protected, and over
the subsequent duration of the experiment 30% ± 11 were removed, a rate much higher than removal from intact exclosures (1.8% ± 0.9) ($F_{1,32.9} = 8.77, P = 0.006$).

When mammals did not have access to $D. cestroides$ seeds, percent seedlings was high and increased through time. Although no germination was recorded through the first 7 weeks, after 63 weeks percent seedlings was 55% ($F_{4,476} = 72.11, P < 0.01$) (Figure 3.3). After 93 weeks, more than 10 times the number of protected seeds had become seedlings (59% ± 6.4) than unprotected seeds (5.7% ± 2.8), mainly due to high rates of removal of the unprotected seeds. Of the 42 unprotected seeds that were not removed 10 became seedlings over the 1.5 years. Despite high removal of seeds when exclosures fell, after 63 weeks the percent seedlings (60% ± 14) in the fallen exclosures was not different from intact exclosures (54% ± 6.3) ($F_{1,113} = 1.29, P = 0.26$). Percent seedlings was not

Figure 3.3 Percent of seeds that were removed or were seedlings, with and without mammal exclosures. The modest drop in removed seeds without exclosures at week 7 resulted from seed replacement in some treatments at week 5.
different between original seeds present at the start of the study and replacement seeds added 5 weeks later ($F_{1,61} = 2.40, P = 0.13$), and there was no significant interaction with census week ($F_{2,122} = 1.78, P = 0.17$).

Neither distance, density, nor any interactions with these variables were important factors in the percentage of seeds removed or in the percent seedlings.

**Fate of Naturally Dispersed Seeds: Predation**

Percent seed predation varied with the interaction of fragmentation and fruit crop years ($F_{1,9.41} = 12.89, P = 0.005$) (Table 3.1). Simple effects tests on this interaction revealed that the 2004 fruit crop had 17 times more seed predation in continuous forest (53% ± 0.5) than in forest fragments (3% ± 0.9) ($F_{1,6.27} = 20.9, P = 0.0003$) but there was no difference for the 2003 fruit crop, when seed predation was very low in both fragments (1.3% ± 0.4) and continuous forest (2.3% ± 0.4) ($F_{1,17.1} = 0.14, P = 0.71$) (Figure 3.4). Seed predation was 23 times higher in continuous forest for the 2004 fruit crop than for the 2003 fruit crop ($F_{1,8.8} = 38.8, P = 0.0002$) but fragments showed no difference in predation between fruit crop years ($F_{1,9.9} = 0.5, P = 0.50$) (Figure 3.4). As noted previously, fruit production in 2004 was much greater than in 2003 (Figure 3.2).

**Fate of Naturally Dispersed Seeds: Seedlings**

Percent seedlings had a significant interaction with all main effects: fragmentation, distance from parent, and time since fruit fall nested in fruit crop year ($F_{16,157} = 1.99, P = 0.02$) (Table 3.1). Simple effects tests on this interaction were used to examine the roles of fragmentation and distance. Under the crown for the 2003 fruit crop the percent seedlings in fragments (28% ± 1.5) was five times that in continuous forest (6% ± 0.28) ($F_{1,32.6} = 4.6, P = 0.04$). Although there were significant distance effects
Table 3.1  Results of the four main effects (F=fragmentation, D=distance, Y=fruit crop year, M=month) and their interactions from fully crossed ANOVAS on seed and seedling fates of naturally dispersed seeds. Bold values represent highest order interactions with significance. Simple effects tests were performed to determine the roles of fragmentation and distance effects on bolded interactions.

<table>
<thead>
<tr>
<th>Fixed Effects</th>
<th>Percent Predation</th>
<th>Percent Seedling</th>
<th>Percent New Seedling</th>
<th>Percent Dead Seedlings</th>
</tr>
</thead>
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<tr>
<td></td>
<td>F</td>
<td>P</td>
<td>F</td>
<td>P</td>
</tr>
<tr>
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<td>0.019</td>
<td>0.2</td>
<td>0.67</td>
</tr>
<tr>
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<td>0.04</td>
</tr>
<tr>
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<td>0.001</td>
<td>3.25</td>
<td>0.06</td>
</tr>
<tr>
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<td>0.04</td>
<td>6.77</td>
<td>&lt;0.0001</td>
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<tr>
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<td>0.0</td>
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</tr>
<tr>
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<td>0.02</td>
</tr>
<tr>
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<td>0.63</td>
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</tr>
<tr>
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<td>0.65</td>
<td>1.99</td>
<td>0.02</td>
</tr>
</tbody>
</table>
Figure 3.4  Percent seed predation (back-transformed means with 95% CI) in continuous forest and forest fragments for the 2003 and 2004 fruit crops. Asterisks indicate significant differences between continuous forest and forest fragments based on simple effects tests.

(Table 3.1), there were no consistent trends associated with fragmentation, distance, or year, so these results are not shown.

Most new seedlings were recorded within 14 months after fruit fall. Percent new seedlings showed a significant interaction between fragmentation, distance, and month nested in fruit crop year ($F_{8,156} = 2.5, P = 0.01$) (Table 3.1), so simple effects tests were used to test for differences based on fragmentation and distance. Shortly after fruit fall, which is in the dry season, a greater percent of seedlings were new in continuous forest than in fragments. Seven months later in the following wet season, this pattern reversed and the percent new seedlings was greater in fragments than in continuous forest (Figure
3.5). This trend is present for the 2002 and 2004 fruit crops, but not the 2003 fruit crop. For the 2002 fruit crop year, this pattern was seen 1 month after fruit fall, whereas it was quicker to develop in 2004 where it was seen immediately after fruit fall (0 months). A two-week delay in the censuses in 2004 can account for the quicker germination of seedlings that year. There were no consistent trends in how distance affected percent new seedlings despite several significant effects (data not shown).

![Figure 3.5](image-url)

**Figure 3.5** The percent new seedlings (back-transformed means with 95% CI) over time since fruit fall for three years. Asterisks indicate significant differences between continuous forest and forest fragments based on simple effects tests ($P \leq 0.01$). Only months with substantial percentages of new seedlings are shown here.

Percent dead seedlings varied only with month nested in fruit crop year ($F_{8,137} = 16.4, P < 0.0001$) (Table 3.1). Dead seedlings were first detected 7 months after fruit fall (16% in 2002 and 12% in 2004), peaked 21 and 24 months after fruit fall, at 23% in 2003 and at 20% in 2002 respectively, and tapered off to 1% by 31 months after fruit fall (2002
only). There were no significant effects of percent dead seedlings with fragmentation or distance effects (data not shown).

Seedling survival and growth were reduced in fragments, especially close to the parent crown. The number of months that seedlings lived varied between fragments and continuous forest and distance class ($F_{2,380} = 3.85, P = 0.05$). In continuous forest seedlings lived longer 0-10 m away from the crown (4 months) than under the crown (2.3 months) or in forest fragments (crown = 2.5 months, 0-10 m = 2 months). Seedling height also varied between fragments and continuous forest and distance class ($F_{2,367} = 4.32, P = 0.01$). Although there was no relationship between seedling height and distance in continuous forest ($F_{2,366} = 1.22, P = 0.30$), seedlings were significantly shorter in fragments than in continuous forest at all distance classes. In fragments, seedlings beyond 10 m from the crown were shorter than under the crown ($P = 0.04$) or 0-10 m beyond the crown ($P = 0.07$) (Figure 3.6).

Although relationships between density and seedling height or survival tended to be significant, given the large sample sizes, the data’s fit to the trend line was poor ($R^2 < 0.01$), so I did not analyze the relationships any further.

**Discussion**

**Fruit Production**

Fruit fall of *D. cestroides* declined as forest fragment size decreased and varied across years in fragments, but not in continuous forest. Lower fruit production in fragments was not a result of smaller trees there as the DBH of fragment trees (77 cm ± 13.2) and continuous forest trees (97 cm ± 14.5) were not different ($F_{1,9} = 1.06, P = 0.33$).
Limited fruit production in fragments could reflect lower pollination in fragments caused by changes in pollinator abundance, density, or behavior (Aizen & Feinsinger 1994, Dick 2001, Powell & Powell 1987). Flowers of *D. cestroides* are yellowish green with long tubular corollas and produce a strong “rose” odor, a morphology indicative of pollination by sphingid moths (Ribeiro *et al.* 1999, Santiago-Valentin & Olmstead 2003). In isolated fragments of South Africa, lower visitation by hawkmoths to individuals of the subcanopy tree, *Oxyanthus pyriformis*, resulted in less than 0.1% fruit set (Johnson *et al.* 2004).

If pollination is not limited, reduced fruit production could reflect resource limitation for fragment trees (Aizen & Feinsinger 1994, Fuchs *et al.* 2003, Horvitz &
Fragmentation causes increased temperatures and decreased humidity (Kapos 1989), perhaps imposing greater physical stresses on some tree species. Where added stress imposes costs, reproductive functions are likely to suffer more than maintenance functions.

**Seed Removal and Predation**

Seed removal and seed predation were high, but differed slightly between the experiment in continuous forest and the naturally dispersed seeds in fragments and continuous forest. In the continuous forest experiment, post-dispersal removal of *D. cestroides* was very high and occurred rapidly, with 72% of seeds removed in the first 5 weeks. However, later in the experiment, removal of the replacement seeds was slower as only 30% of them were removed between weeks 5 and 93. Similarly, removal of seeds from the exclosures that collapsed between weeks 7 and 63 was also 30%. I do not know whether seed removal represented secondary dispersal or seed predation in this experiment. However, for the naturally dispersed seeds, I monitored predation, which was over 53% of the seeds in continuous forest versus only 3.1% in fragments for the 2004 fruit crop. In 2003, the fruit crop was exceptionally small, and predation was very low in fragments and continuous forest.

I hypothesized that there would be a difference in seed predation between fragments and continuous forest but I did not predict the direction of the difference because fragmentation has produced variable effects on different mammal species at the BDFFP. Malcolm (1997) clearly showed that fragments have larger populations of small mammals, mostly rats and opossums. However, Malcolm’s studies did not include larger mammals such as agoutis (*Dasyprocta sp.*), acouchies (*Myoprocta sp.*), peccaries and the
Guianan squirrel (*Sciurus aestuans*). Acouchies (M. Jorge per. comm.), agoutis, white-lipped and collared peccaries, and pacas at the BDFFP have higher densities or sightings in continuous forest than in fragments (Timo 2003). Such medium-sized rodents may operate as seed predators but they are also secondary seed dispersers.

Few data exist on which animals are predators or secondary dispersers of *D. cestroides* seeds. Terrestrial camera traps at three continuous forest sites have recorded interest in *D. cestroides* fruit by the Guianan squirrel (*Sciurus aestuans*), brown four-eyed opossum (*Metachirus nudicaudatus*), a species of mouse or slender mouse opossum (*Marmosa* or *Marmosops* spp.), a margay (*Leopardis wiedii*), grey-winged trumpeters (*Psophia crepitans*), agoutis (*Dasyprocta* sp.), and a capybara (*Hydrochaeris hydrochaeris*) (Yabe et al. 1998, Ch. 2). In addition, acouchies are hypothesized to be dispersers (Ribeiro et al. 1999), and I speculate that pacas, peccaries, and rats may also be important for *Duckeodendron* dispersal and/or predation. Evidence of buried seeds and clusters of predated seeds in the field support the notion that medium-sized, scatter-hoarders, like agoutis and acouchies, may be the most important seed predators or secondary-dispersers for *D. cestroides*.

In this regard, it is noteworthy that the percent of seeds removed in the experimental study in continuous forest (77% after 93 weeks) exceeded the percent of seed predation in my observations of naturally dispersed seeds in continuous forest when seed predation was high there (53% in 2004). This difference between removal and predation rates may represent secondary dispersal, as removal from the experiment was an additive effect of dispersal and predation. The role of agoutis as seed dispersers has been demonstrated for some palm species because only scatter-hoarded (buried) seeds escape predation (Brewer...
et al. 2000). Therefore, fragmentation effects on medium-sized rodents may not only
decrease seed predation, but also secondary dispersal and scatter-hoarding, which will
reduce *D. cestroides* seed survival.

**Seedlings**

Escaping mammalian seed predators confers a huge advantage for seedling
germination. When protected from mammals, *D. cestroides* seeds in continuous forest
have a high probability of becoming seedlings. Naturally dispersed seeds were less likely
to establish as seedlings (15%) than experimentally protected ones (59%), probably
because rodent seed predation is so high. In 2003, when seed predation was very low, I
observed the highest rates of percent seedlings among naturally dispersed seeds – 28% in
fragments and 6% in continuous forest; however, these values still do not approach the
rates of seedling establishment for protected seeds. Even seeds protected from predators
for just the first few weeks after fruit fall, as in cases of fallen exclosures, have a 3.5 to
9.5 times greater likelihood of becoming seedlings (54%) than naturally dispersed (15%)
or unprotected seeds (5.7%). Fallen exclosures were not detrimental to seedling
establishment because they protected seeds until they germinated, generally within the
first month in continuous forest (Figure 3.5). While recorded percentages of seedlings
were high, germination may have been higher as I could not document seeds that
germinated and died between censuses, especially for the later censuses at weeks 63 and
93.

Differences in seed germination rates may result from microclimate variation
between fragments or continuous forest. Most naturally dispersed seeds in continuous
forest germinated within the first few months after fruit fall whereas germination in fragments was delayed several months. This delay in germination in fragments coincides with Central Amazon’s seasonal shift from the dry to the wet season. *Duckeodendron* fruits fall between June and August, the three driest months of the year with mean monthly rainfall just above 100 mm (Laurance 2001). Seven months later, March and April have more than twice the rainfall of the dry season months (Laurance 2001). In fragments where there is decreased humidity and soil moisture and increased temperatures in the understory (Kapos 1989), seeds may need moisture from the rainy season to germinate.

Five times the percent of seeds became seedlings under the crown in fragments than in continuous forest. Why did so many seedlings establish under the crown only? First, reduced seed dispersal in fragments (Ch. 2) results in the majority of seeds concentrated close to and under the parent crown. Second, low levels of seed predation in fragments results in more seeds that can become seedlings. In contrast, in continuous forest more seeds are dispersed away from the crown and seed predation is higher, leaving few seeds under the crown to establish as seedlings. This effect might have been the strongest in 2003, when fruit production was lowest because seed dispersal distances, even in fragments, are affected by overall fruit availability with lower dispersal distances in years with lowest fruit production rates (Ch. 2). Therefore, when fruit production is low, fewer seed dispersers exaggerate the tendency of seeds in fragments to fall directly below parent crowns and result in the high percent of seedlings seen there.

I expected that processes of density- and distance-dependent mortality would be more exaggerated in fragments than in continuous forest. In continuous forest,
manipulated seed densities at three distances from parent crowns showed no effects of
density or distance on mammal seed removal or seedling establishment. All unprotected
seeds were rapidly removed, even solitary seeds 35 m from the parent crown. Likewise,
seeds even in high densities close to parent crowns showed high seedling establishment.

Similarly, seedlings from naturally dispersed seeds showed no evidence of distance-
dependent mortality. Although there were differences among distance classes for percent
seedlings and the percent of new seedlings, there were no consistent trends in how these
varied with fragmentation, time since fruit fall, and fruit crop year. The lack of distance
or density-dependent effects on percent seedlings indicate no Janzen-Connell processes at
the seedling stages I measured.

Distance- and density-dependent mortality can be influenced by rodent and insect
seed predators. I identified only seed predation by rodents and not insects as seeds were
monitored for the entire length of the study and could not be opened. Predation by
insects, such as bruchid beetles, is often severe among tropical seeds, although the effects
of fragmentation on insect predation are mixed (Cascante et al. 2002, Chacoff et al. 2004,
Wright & Duber 2001). Over several years of observing seeds in the field, I never saw
any exit holes indicating insect predation for D. cestroides. However, ants were
sometimes observed emerging from the endocarp and various insects, including beetles,
were sometimes seen among the fibers of the mesocarp. Sometimes differential effects of
rodent and insect predation in fragments and continuous forest combine to result in
similar total predation rates across forest sizes (Wright & Duber 2001). Higher insect
predation in fragments could explain the lack of differences in seedling establishment in
fragments and continuous forest seen here.
Seedling Survival and Growth

Increased seedling mortality has been cited as one of two major factors that could reduce seedling densities in fragments (Benitez-Malvido 2001) but I found no evidence of higher mortality in fragments than in continuous forest. Although seedlings in fragments lived fewer months than in continuous forest, this result is a consequence of the delayed germination of seedlings in fragments and my recording the number of months seedlings lived only until the termination of the experiment. I also showed that seedlings in fragments were only half as tall as their counterparts in continuous forest, perhaps another consequence of the delayed germination. However, this consequence may be more than an artifact, because the seedlings are different heights but from the same cohort of seeds. Larger seedlings commonly have higher survival rates than smaller seedlings, especially in seasonal droughts (Gilbert et al. 2001). Therefore shorter seedlings in fragments may have reduced survival, primarily because of delayed germination.

This study has two obvious limitations. First, I did not account for fruits and seeds removed and deposited outside or beyond transects. Increased fruit removal and dispersal beyond transects and traps would result in lower counts of fruit in sampled areas and could falsely indicate low fruit production. My methods do not account for rare long-distance dispersal events that involve just a few fruits, which might be crucial to seedling establishment and recruitment. These seeds were beyond the scope of this study.

The second limitation concerns non-parallel censuses of naturally dispersed seeds among fruit crop years. The lack of replicated censuses at all time intervals across the
three years makes it difficult to draw conclusions about changes in seed fate over time. Primarily for this reason I had few \textit{a priori} hypotheses concerning time since fruit fall. However, when month proved important, as in the appearance of new seedlings in fragments and continuous forest, the overall effect was apparent for several years.

\textbf{Conclusions}

This study provides strong evidence that fruit production and post-dispersal seed fate of \textit{D. cestroides} are altered in forest fragments. Fragment trees have lower fruit fall, thereby contributing fewer seeds for regeneration. Although seed predation was much higher in continuous forest than in fragments, this may also be associated with higher rates of secondary dispersal and seed caching, which confer an advantage for seedling establishment. With the addition of reduced quantities and distances of dispersed seeds in fragments (Ch. 2), it seems that regeneration of \textit{D. cestroides} is at risk from multiple factors. I found no discernable differences in the percent of seeds that became seedlings or the percent of seedlings that died in fragments and continuous forest for the first few years after fruit fall. It is difficult to predict how dramatic shifts in seed predation and delayed germination will impact seedlings over the long term. However, dramatic changes in seedlings between fragments and continuous forest may surface over the long term, especially as many of the processes that influence seedling establishment, i.e., seed dispersal and seed predation, are altered by fragmentation. Such pervasive changes may limit \textit{D. cestroides'} ability to persist in fragments.
CHAPTER 4

FOREST FRAGMENTATION DIFFERENTIALLY AFFECTS SEED DISPERsal OF LARGE AND SMALL-SEEDED TROPICAL TREES

Introduction

Responses of mutualisms to forest fragmentation can be very diverse. Declines in animal or plant species can result in the loss of mutualistic functions, jeopardizing connected species (Andresen & Levey 2004, Bruna et al. 2005). Alternatively, some mutualisms are more resilient to fragmentation because the behavior of animal mutualists is not altered, lost mutualists are replaced by other species (Dick 2001), or lost mutualistic functions are replaced by the plant itself through self-fertilization (Powell & Powell 1987). Given the diversity of individual responses to fragmentation, are there generalizations that allow us to predict the effects of forest fragmentation for some tropical rainforest trees?

It has been hypothesized that populations of large-seeded plants will be highly susceptible to fragmentation (Fortuna & Bascompte 2006, Jones & Crome 1990, Kitamura et al. 2004, Melo et al. 2006, Silva & Tabarelli 2000, Tabarelli & Peres 2002). In contrast, small seeded plants may sustain viable populations in or across forest fragments (Silva & Tabarelli 2000, Tabarelli & Peres 2002). Seed dispersal of large and small-seeded tree species may be differentially affected by forest fragmentation because their dispersers have varying responses to fragmentation for several reasons. First, large-seeded species rely on fewer animal species for dispersal (Christian 2001, Hamann & Curio 1999). Second, dispersal agents of large-seeded species are often larger and therefore at greater risk of extirpation in fragments, given greater hunting pressures, their low reproductive rates, and small populations (Bodmer et al. 1997, Gilbert & Setz 2001, Hamann & Curio 1999, Willis 1979). In addition, large-animals cannot subsist even in large fragments (100 ha) because their home ranges are too extensive (Chapman 1989,
Woodroffe & Ginsberg 1998). After forest fragmentation, the loss of the few dispersal agents available for large-seeded plants can effectively eliminate all seed dispersal. On the other hand, small-seeded fruits can be dispersed by many species of frugivores and extinction of any single disperser might have little effect on a tree’s net dispersal ability (Corlett 1998). The added vegetative diversity provided by edge habitats and the matrix around fragments attracts small, omnivorous frugivores (Galetti et al. 2003, Pizo 1997), potentially increasing the dispersal community for small-seeded species (Blake & Loiselle 2001, Jules & Shahani 2003).

Here, I offer a preliminary test of the hypothesis that forest fragmentation negatively impacts seed dispersal of a large-seeded tree species more than that of a small-seeded tree species. I examined the effects of forest fragmentation on the seed dispersal of two Amazonian tree species in a year of high fruit production. *Duckeodendron cestroides* Kuhlm. (Solanaceae) is a Central Amazon endemic with large fruits (5.9 cm long). *Bocageopsis multiflora* Mart. (Annonaceae) is more widely distributed and has small fleshy berries (0.8 cm diameter).

This study is the first comparison of fragmentation effects on the seed dispersal of two coexisting tree species with different fruit sizes in the aseasonal tropics. As such, I confronted several challenges: individuals of most species are rare (Oliveira 1997), tree species fruit supra-annually usually with different fruiting phenologies; furthermore, measuring dispersal of large and small-seeded species requires different sampling methods (Herrera et al. 1998). Results are tempered accordingly.
Methods

Study Site

I sampled trees within the experimentally fragmented reserves of the Biological Dynamics of Forest Fragments Project (BDFFP), a collaboration between Brazil’s Institute for Amazonian Research (INPA) and the Smithsonian Tropical Research Institute (STRI). The BDFFP reserves are located north of Manaus at 2°30’ S and 60°00 W in the State of Amazonas Brazil. Vegetation is upland terra firme tropical moist forest with an elevation range of 50-100 m. Mean annual rainfall is 2,651 mm with a mild dry season from June to October. The BDFFP has 11 isolated reserves distributed in 1-ha, 10-ha, 100-ha forest fragments and control plots in nearby continuous forest, established between 1980 and 1984. Each reserve includes at least one hectare where all trees ≥ 10 cm DBH (diameter at breast height) are censused. This database of trees allowed me to identify and locate individuals of our target species from over 400,000 living, tagged trees across 66 hectares of tropical forest.

Study Species

I identified one large-seeded species and one small-seeded species, based on fruit crops that were large enough to sample. As the two species exhibited different fruiting phenologies, they were sampled during consecutive dry and rainy seasons.

Duckeodendron cestroides (Solanaceae) is a large-seeded emergent tree endemic to the Central Amazon. It is one of the largest trees in the area with a maximal DBH of 153.2 cm (Laurance et al. 2004). Adults are rare at the BDFFP with recorded densities of 1.3/ha (Oliveira 1997) and 0.5/ha (BDFFP database). In the dry season trees produce narrowly ovate fruits that average 5.9 by 3.4 cm in size and 26.8 g fresh weight (n=50).
Pulp of *D. cestroides* is very thin, so fruit and seed sizes are about the same. When ripe, fruit pulp is orange-yellow with a strong, musty odor. *Duckeodendron* has been recorded in the diets of two primate species – red howler monkeys (*Alouatta seniculus*) (Neves & Rylands 1991, P. Stouffer, pers. obs.) and bearded sakis (*Chiropotes sagulatus*) (S. Boyle, pers. obs.). Once fruit has fallen, terrestrial mammals may be important dispersers (Yabe *et al.* 1998, Ch. 2).

The small-seeded species, *Bocageopsis multiflora* (Annonaceae), is a canopy tree that grows maximally to 33.1 cm DBH (Laurance *et al.* 2004). This species is also rare locally, BDFFP adult densities: 3.7/ha (Oliveira 1997) and 2.3/ha (BDFFP database), although widely distributed throughout the Brazilian Amazon and Guianas (Webber & Gottsberger 1995). In the wet season, between February and May (Oliveira 1997), trees produce ripe fruits that are 0.8 cm in diameter, with a watery orange to purple pulp around 1-2 seeds. Seed size is 0.5 cm diameter when there is one seed per fruit, and about half this size when there are two seeds. Fruit size and color implicate frugivorous birds of all sizes as consumers of this species, and seeds have been found in bird feces (M. Gordo, pers. obs.).

Fruit and seed sizes of *D. cestroides* and *B. multiflora* are representative, respectively, of large- and small-seeded tropical species (Hammond & Brown 1995). Pantropically, fruit size among the eight largest families that rely on animals for dispersal ranges from 0.5 to 15 cm long (Mack 1993). In the Neotropics, the smallest seeds, less than 0.1 cm long or 0.001 g wet weight, come from figs (*Ficus* spp.) and the Melastomataceae. Generalized, frugivorous birds disperse small seeds (up to 1 cm long), like *B. multiflora* (Howe & Westley 1986). In contrast, mammals disperse seeds greater
than 5 cm long (Howe & Westley 1986) with an average wet weight of 12.6 ± 2.5 g (Hammond & Brown 1995). *Duckeodendron* seeds, 5.9 cm long and 26.8 g wet weight, are among the largest seeds and fruits in Neotropical forests.

**Seed Dispersal**

Given their different seed sizes, I sampled dispersal differently for the two species, trying to employ parallel methodologies. The large *D. cestroides* produces a small number of large fruits whose seeds persist on the ground many months after dispersal, so I located them in the leaf litter. *B. multiflora* seeds are small and impossible to census in the leaf litter, so I sampled them using seed traps. For both species, the area censused or trapped increased with distance from the parent tree to compensate for decreased seed densities (Clark et al. 2005, Ch. 2).

*Duckeodendron* seed dispersal was censused during the dry season of 2002 (June-Oct.) using 11 fruiting adults; four in 10-ha fragments, two in 100-ha fragments, and five in continuous forest. I established two wedge-shaped transects originating at the center of each focal tree’s canopy in 2002 (Ch. 2). Each transect had a 45° base angle and together the two transects represented 25% of the tree’s potential dispersal area for censused distances. I counted all seeds within these transects and measured distances, to the nearest meter, from the edge of maternal tree crowns. Transects were terminated after three to four consecutive distance classes revealed no seeds. See Chapter 2 for a more detailed description of transect construction and placement.

In the following wet season (Feb.-April 2003), I identified eight fruiting adults of *B. multiflora* for study: one in a 1-ha fragment, one in a 10-ha fragment, two in 100-ha fragments, and two each in two continuous forest sites. I placed seed traps around *B.*
multiflora individuals along a single line-transect originating from the center of the crown of each focal tree. As with D. cestroides, these transects were oriented to avoid dispersal overlap from other fruiting conspecifics, fragment edges, trails, or plot edges where tree composition was unknown. The tree in the 1-ha fragment was virtually in the center of its fragment where edge effects are less severe. Ten seed traps were mounted along each transect: one trap each mid-way between crown center and crown edge, at 5 m and 10 m from the crown edge; two traps each at 20 m and 35 m from the crown edge, and three traps at 50 m from the crown edge. I chose not to place traps where there would be unusually low volumes of seeds such as directly underneath palm fronds or other low branches that would deflect seeds, or in gaps where there were no available perches for bird dispersers.

Seed trap frames were constructed with 0.71 m long pieces of 1/2” PVC, creating a 0.5 m² trapping area. To reduce seed predation, traps were raised 0.5 m off the ground. Along three points of each side of the PVC frame I secured fiberglass mesh screen (18x16 squares per in²) with 0.11-inch diameter wire (Phifer Wire Productions, Tuscaloosa, USA). The screen was fastened so that it created a sagging net that would catch and cushion falling seeds. I removed and counted all intact and partial fruit, and seeds from each trap biweekly until fruiting and dispersal ended. Seeds that were clean with no remaining pulp were considered dispersed; all seeds found in traps beyond the crown were clean.

**Statistical Analyses**

To test for an effect of fragment sizes on seed dispersal and the validity of lumping all fragment sizes together, I ran a generalized linear mixed model (PROC GLIMIX)
with a Poisson distribution on the percentage of seeds dispersed more than 1 m past the crown for each species. This analysis revealed no difference in dispersal between fragment sizes for *Duckeodendron* \( F_{1,4} = 0.68, P = 0.46 \) and for *Bocageopsis* \( F_{2,1} = 2.7, P = 0.40 \). Therefore, for each species I grouped trees from all fragment sizes as fragment trees \( n=6 \) for *Duckeodendron*, \( 4 \) for *Bocageopsis* and compared them to continuous forest trees \( n=5 \) for *Duckeodendron*, \( 4 \) for *Bocageopsis* in all analyses.

To test for effects of forest fragmentation on the seed dispersal of each species I performed three types of analyses. The first was a generalized linear mixed model (GLMM) with a binomial distribution and a logit transformation to examine the percentage of seeds that were dispersed more than 1 m beyond the crown. The second was a set of GLMMs to test the effect of fragmentation on dispersal distances including 1) the mean distance of all dispersed seeds, 2) the mean distance of the five furthest dispersed seeds for each tree (Poisson distribution and log transformation), and 3) the number of seeds dispersed more than 10 m beyond parent crowns (Poisson distribution and log transformation). Each of these analyses included fixed effects of forest type (fragments and continuous forest) and random effects of individual trees. The third type of analysis was a two-sided Random Coefficient Regression Analysis (RCR) with random intercepts only, and assuming a Poisson response to compare distributions of dispersed seeds across distances censused. An RCR is a generalized linear mixed model that fits subject-specific models about population-averaged models, similar to an analysis of covariance except that it assumes random effects for the covariate (Littell *et al.* 1996, Moser 2004). I natural log-transformed both seed number and dispersal distance.
variables for regressions used in the RCR analysis for Bocageopsis \((R^2 = 0.20 - 0.82, 4\) of 8 values significant to \(P = 0.05\)), but regressions using untransformed values for seed number provided better fits for Duckeodendron \((R^2 = 0.40 - 0.81, 8\) of 11 values significant to \(P = 0.05\)). Offsets of total number of fruits and sample area (natural log-transformed), were included for the percentage of dispersed seeds and the RCR analysis, respectively, to accommodate each tree’s differentially sized fruit crops and sampling areas. Response variables were divided by the offset, transforming them into proportions. The fractional degrees of freedom seen throughout the paper are a result of a Kenward-Rogers adjustment to the degrees of freedom, which provides a conservative Type I error control for small sample sizes. All analyses were performed with the SAS System for Windows (version 9.1) (SAS Institute Inc. 2002).

**Results**

A total of 1,805 *D. cestroides* seeds were counted in the dry season of 2002. A total of 1,367 *B. multiflora* fruits were captured in seed traps, of which 396 were classified as dispersed seeds. Seed fall in continuous forest was not different than in fragments for *D. cestroides* (continuous = 196, fragment =84, \(F_{1,9} = 4.07, P = 0.07\)) or *B. multiflora* (continuous = 201, fragment = 67, \(F_{1,6} = 3.5, P = 0.11\), dependent variable natural-log transformed for normality), although I still controlled for differential fruit production across individuals by including total fruit fall as an offset variable in the analysis that included dispersed and undispersed seeds. Likewise, large trees might produce more fruit than small trees but the DBH of our trees in continuous forest (*D. cestroides* \(= 97 \text{ cm} \pm 14.5, B. multiflora = 31 \text{ cm} \pm 1.8\)) was not different from the DBH in forest fragments for
either species (*D. cestroides* = 77 cm ± 13.2, *B. multiflora* = 33 ± 1.8) (*D. cestroides*, $F_{1,9}$ = 1.06, $P = 0.33$, *B. multiflora* $F_{1,6} = 0.52$, $P = 0.50$).

All analyses indicated a difference in dispersal between continuous forest and forest fragments for *D. cestroides*, but none indicated a difference for *B. multiflora*. In continuous forest 48% ± 6 of *D. cestroides* seeds were dispersed more than 1 m past parent crowns whereas in fragments only 16% ± 3 of seeds were dispersed that far ($F_{1,7} = 23.2, P = 0.002$; Figure 4.1A). In addition, *D. cestroides* seeds were dispersed one and a half times further in continuous forest (mean = 7.8 m ± 1) than in forest fragments (mean = 3.0 m ± 0.9) ($F_{1,9} = 12.22, P = 0.007$; Figure 4.1B). The 5 furthest dispersed seeds of *D. cestroides* were dispersed 5 times further in continuous forest (mean = 20.5 ± 6.7) than in forest fragments (mean = 4.4 ± 1.5; $F_{1,8} = 10.41, P = 0.01$; Figure 4.1C) and many more seeds per tree were dispersed 10 m beyond the crown in continuous forest (mean = 24.7 ± 19) than in forest fragments (mean = 0.4 ± 0.4; $F_{1,8.3} = 11.02, P = 0.01$; Figure 4.1D).

In contrast, for *B. multiflora* there was no difference between continuous forest and fragments in either the percentage of dispersed seeds (continuous forest = 5% ± 5, fragments = 12% ± 10, $F_{1,5} = 0.47, P = 0.52$), mean distance of dispersed seeds (continuous forest = 23.1 m ± 8.0, fragments = 18.6 m ± 8.0, $F_{1,6} = 0.15, P = 0.71$), distance of the 5 furthest dispersed seeds (continuous forest = 16.2 ± 10.7, fragments = 17.6 ± 11.2, $F_{1,4} = 0.01, P = 0.93$), or number of seeds dispersed more than 10 m (continuous forest = 7.2 ± 4.3, fragments = 5.0 ± 3.0, $F_{1,4.1} = 0.2, P = 0.68$) (Figure 4.1).

Although distributions of dispersed seeds in continuous forest and forest fragments were non-linear and highly variable among individuals of both species, Random
Figure 4.1  Seed dispersal for each species in continuous forest and forest fragments.  A) Percentage of seeds dispersed more than 1 m past the crown.  B) Mean distance dispersed seeds were found from the crown of each tree.  C) Mean dispersal distance of the five furthest dispersed seeds for each tree.  D) Number of seeds dispersed more than 10 m beyond the crown of each tree.  Graphed values represent LSMeans with 95% CI from 1-way ANOVAs of each species.  Asterisks indicate significant differences between fragments and continuous forest.
Coefficient Regressions showed that the number of seeds dispersed per 10 m$^2$ declined as distance from the parent crown increased for both species ($D. cestroides F_{1,9.4} = 6, P < 0.0001$, $B. multiflora F_{1,26.8} = 4.57, P = 0.04$). The slopes of the regressions were not different in fragments versus continuous forest for either species ($D. cestroides F_{1,85} = 0.66, P = 0.41$, $B. multiflora F_{1,26.8} = 0.80, P = 0.32$). However, intercepts were significantly lower in forest fragments than in continuous forest for $D. cestroides (F_{1,9.4} = 6 P = 0.035)$, indicating that more seeds were dispersed to all distances beyond the crown in continuous forest than in forest fragments (Figure 4.2). In contrast, intercepts were not different between continuous forest and fragments for $B. multiflora (F_{1,33} = 0.58, P = 0.45)$, indicating that seed dispersal was distributed similarly across distances for both continuous forest and forest fragments (Figure 4.2).

**Discussion**

These results demonstrate that forest fragmentation does not alter the seed dispersal of the small-seeded $B. multiflora$, but dramatically reduces the seed dispersal of the large-seeded $D. cestroides$. For $D. cestroides$ there is a threefold reduction in the percent of seeds dispersed and the mean distance of dispersed seeds in forest fragments. More dramatic results were that each tree’s five furthest dispersed seeds were five times farther from the crown in continuous forest and that fifty times more seeds were dispersed 10 m beyond the crown in continuous forest. Finally, distributions of dispersed seeds across distances from the parent tree also showed fewer seeds at all distances.

I attribute the differences in seed dispersal of $D. cestroides$ and $B. multiflora$ to the differential effects of forest fragmentation on the dispersal guilds of small and large-seeded species. Effective dispersers for large-seeded species like $D. cestroides$ are limited to medium
Figure 4.2  Predicted density of dispersed seeds per 10 m² (solid lines) and 95% confidence intervals (dotted lines) across distances from maternal trees in fragments and in continuous forest for *D. cestroides* and *B. multiflora*. For visual clarity, the X-axis of *D. cestroides* graph is abbreviated.
and large mammals because smaller animals are simply incapable of handling large fruits (Jordano & Schupp 2000, Wheelwright 1985) whereas disperser size is not as limiting for small fruits (Dowsett-Lemaire 1988, Kitamura et al. 2004, Wheelwright 1985). The average number of dispersers in a Philippine submontane rainforest was twice as great for species with fruits < 20 mm (8.5 dispersers N = 13) as for species with fruits > 20 mm (4.8 dispersers N = 11) (Hamann & Curio 1999). In a survey of the Oriental Region, Corlett (1998) found that fruits with a diameter larger than 30 mm can only be consumed by the ecosystem’s largest frugivores — hornbills, fruit pigeons, and fruit-eating mammals.

Large-seeded species also tend to be highly specialized with respect to their dispersers. In Peru, the large-seeded Virola calophylla (mean seed length = 17 mm) relies on spider monkeys (Ateles paniscus body length = 43-62 cm) to disperse 92% of its seeds (Russo 2003). Similarly, in Thailand, virtually 100% of seed dispersal of the large-seeded Aglaia spectabilis (seed length = 3.0 - 4.6 cm) was accomplished by four species of hornbill, of which only one (Buceros bicornis body length = 120 cm), accounted for 65% of dispersal (Kitamura et al. 2004). Finally, in a Philippine rainforest three tree species depend solely on two species of hornbill (Aceros waldeni and Penelopides panini) for seed dispersal and two other species depend entirely on the visayan warty pig (Sus cebifrons) for seed dispersal (Hamann & Curio 1999).

In addition to large-seeded trees being specialized to their dispersers, these dispersers are often specialized on their plant species. Specialist frugivores are usually large (> 250 g), depend on fruit for a critical portion of their diet, and exist in small populations (Howe 1993). Primates, large terrestrial mammals, and large birds are commonly considered specialist frugivores. In contrast, small-seeded trees are more reliant on generalist dispersers, including passerines and frugivorous bats. Generalist frugivores are smaller (< 50 g), supplement their diet with fruit, and
exist in large populations (Howe 1993). Specialized interactions in dispersal mutualisms are uncommon, making them more susceptible to fragmentation, whereas weak generalist interactions are abundant, making them more resilient to fragmentation effects (Bascompte et al. 2006).

At the BDFFP responses of primates and large mammals to fragmentation mirror the changes in seed dispersal of D. cestroides. More than six times the number of medium and large terrestrial mammals were recorded in fragments than in continuous forest, with especially large differences for brocket deer (Mazama spp.), white-lipped peccaries (Tayassu pecari), collared peccaries (Tayassu tajacu), and agoutis (Dasyprocta leporina) (Timo 2003). Half of the BDFFP’s primate species have disappeared from fragments (Gilbert & Setz 2001, Rylands & Keuroghlian 1988). In contrast, many guilds of frugivorous birds did not suffer declines in density following forest fragmentation (Stouffer et al. 2006), suggesting that entire guilds or some species within each guild were resistant to fragmentation. Remaining species of frugivorous birds are the current dispersers of B. multiflora in fragments.

The scope, length, and methods of our study each limit my interpretations about seed dispersal. First, seed dispersal among individuals of the same species and within the same forest type were highly variable. One D. cestroides individual in continuous forest had a seed dispersal distribution more similar to fragment trees than other continuous forest trees. B. multiflora trees in both fragments and continuous forest had anywhere from 0% to 40% of their seeds dispersed. This variability may reflect the probabilistic character of seed dispersal, such that the effects of fragmentation appear in a non-deterministic fashion.

Second, our study was limited to one-year and cannot account for effects of supra-annual variation in fruit abundance, although I do know that fruit production was high in the year of our
study. Fruit production can directly impact disperser visitation which may be important to overall seed dispersal. Multiple years of sampling might show that the lack of differences in dispersal between fragments and continuous forest for *B. multiflora* was a result of annual variation in disperser abundance or behavior, not fragmentation.

Finally, our sampling methods impose some limits on the interpretation of our results. Although the design of our study limits the certainty of each seed’s parental origin, the isolation of each individual from fruiting conspecifics limits problems associated with parental origin (Ch. 2). Also, our study cannot account for long-distance seed dispersal, although I believe that my samples are representative of seed dispersal for each tree (Ch. 2). Furthermore, sampling one species with seed traps and the other by counting seeds on the forest floor opens the possibility of methods affecting the results. However, the nature of this study, comparing two species with contrasting seed sizes, precluded using the exact same methodology and the exact same sampling months.

With these limitations I achieved a modest comparison of a single large and a single small-seeded species from over a thousand species in the BDFFP forests (Gascon & Bierregaard 2001). Thus, I am unable to generalize our results to differences between all large and small-seeded fruits. However, my data in conjunction with other research on fragmentation support the hypothesis tested.

**Evidence From Tropical Fragmentation Literature**

Three lines of evidence from research on disperser responses to fragmentation support the hypothesis that, in the tropics, the dispersal of large-seeded tree species is more threatened by forest fragmentation than the dispersal of small-seeded tree species. First, large-seeded species are prone to extinction, especially in fragments, because they depend on large dispersers that
require large home ranges and are susceptible to edge effects and hunting. The extinction of the four largest frugivorous pigeons in Tonga (Polynesia) left 18 large-seeded plant species with no dispersers (Meehan et al. 2002). In Madagascar the extinction of 16 large-bodied mammals left the collared brown lemur (*Eulemur fulvus collaris*) as the sole disperser for five large-seeded species (Bollen et al. 2004b). In the highly fragmented Atlantic forest of Northeastern Brazil, the largest frugivorous bird (*Mitu mitu*) is extinct, and large-bodied frugivores are the most vulnerable to extirpation as a result of fragmentation (Silva & Tabarelli 2000, Tabarelli & Peres 2002). In Ugandan forests, the loss of elephants has left *Balanites wilsoniana* regeneration concentrated under parent plants (Babweteera et al. 2007). At the BDFFP, primates are less common or often absent from fragments (Gilbert & Setz 2001). Highly frugivorous primates, such as *Ateles*, are found only in large tracts of forest whereas more folivorous primates, such as *Alouatta*, still occur in forest fragments (Estrada & Coates-Estrada 1996). In forest remnants in Singapore and Hong Kong, entire fruit crops remain underneath parent crowns of large-seeded trees, evidence that their dispersers are absent from these fragments (Corlett & Turner 1997).

In contrast, dispersal of small seeds may remain unaltered by fragmentation because their dispersers are unaffected by fragmentation. In the Atlantic Forest of Brazil, fragmentation reduced the number of bird visitors and effective dispersers for the small-seeded *Cabralea canjerana* (mean size = 4 mm/diaspore) (Pizo 1997). However, there was no indication that the residual guild of dispersers at the fragmented site did not continue to provide effective seed dispersal for *C. canjerana*. Bird assemblages visiting *Dendropanax arboreus* (fruit length = 4-7 mm) in Mexico were different between a 650-ha forest reserve and riparian remnants, but these differences did not translate into different fruit removal rates (Graham et al. 2002).
The second line of evidence is that forest fragmentation restricts or alters the movement of large dispersers more than small ones. Primates seldom move between fragments, especially when patches are devoid of preferred food sources (Bollen et al. 2004a, Estrada et al. 1994, Gascon et al. 2002, Gilbert & Setz 2001, Ratiarison & Forget 2005, Schwarzkopf & Rylands 1989). Movement of frugivorous bats and small omnivorous birds is not as disrupted by forest fragmentation (Corlett 1998). Bats are important dispersers in disrupted landscapes and often contribute the majority of dispersed seeds across fragments or in the regenerating matrix (Bollen et al. 2004a, Gorchov et al. 1993, Parrotta et al. 1997). Avian dispersers are very mobile and track fruit resources among rainforest patches (Price 2004) even venturing into open habitats when perches are available (Shiels & Walker 2003).

Finally, there is some evidence that large and small-seeded fruits, respectively aligned with specialist and generalist dispersers, are differentially linked with primary versus secondary forests in the tropics. Pioneer plant species have smaller seed masses than mature forest species (Foster & Janson 1985, Richards 1996) and large-seeded species are less represented in regenerating forest than in primary forest (Parrotta et al. 1997). Mammal dispersed trees (excluding bats) are more abundant in late-successional habitats than early-successional habitats (Hamann & Curio 1999). On the other hand, recently cleared and early successional habitats that characterize edge communities and small fragments are composed of wind, generalist bird and bat dispersed seeds (Foster et al. 1986, Gorchov et al. 1993, Ingle 2003, Medellín & Gaona 1999, Parrotta et al. 1997). In fact, 82% of seeds larger than 15 mm were only recorded in the interior of a 3500 ha fragment (Melo et al. 2006). In contrast, at forest edges, species with seeds smaller than 6 mm were more abundant in the seed rain (Melo et al. 2006) and small artificial fruits were visited by birds more frequently than in forest interiors (Galetti et al. 2003). In the
fragmented Atlantic Forest, the percentage and number of small-seeded (< 0.6 cm) species were reduced as the successional age of plots increased, while the percentage and number of medium-sized (0.6 – 1.5 cm) species increased with plot age (Tabarelli & Peres 2002). At the BDFFP, genera that decline in fragments were characterized by many factors including large seeds and later successional status (Laurance et al. 2006a). In disturbed habitats, remaining populations of generalist disperser assemblages are likely to consist of species with partially omnivorous diets that do well in early to mid-successional habitats (Shiels & Walker 2003) while large specialist dispersers are reluctant to cross open habitats. Therefore, although large-seeded species could succeed in early-successional habitats, their large size limits the likelihood of their arrival there (Foster et al. 1986, Wunderle 1997). In a fragmented Kenyan forest, pioneer plant species were abundant in the seedling community but 75% of non-pioneer species showed reduced regeneration (Githiru et al. 2002a).

How do these different patterns of dispersal for early and late-successional species relate to forest fragmentation? At the BDFFP, there is clear evidence that forest fragments accumulate secondary forest trees in deference to primary forest species (Laurance et al. 2006b). We found *D. cestroides* only in continuous forest or the core of 10 and 100-ha forest fragments. On the other hand, *B. multiflora* can be found throughout the forest fragments and in secondary forests, indicating that its dispersers not only move between fragments, but also frequent the regenerating matrix around fragments.

Other studies on seed dispersal in forest fragments support the hypothesis that dispersal of small-seeded species is less affected by fragmentation than large-seeded species. Nine small-seeded tree species (<14 mm) in southeastern Kenya showed high fruit consumption in forest fragments, although there was low disperser mobility between fragments (Githiru et al. 2002b).
Second, fewer dispersal agents and reduced seed dispersal were detected in fragments compared with continuous forest for the endemic large-seeded *Leptonychia usambarensis* (seed size = 90-133 mm) in Tanzania (Cordeiro & Howe 2003). Two other studies have examined frugivore assemblages in isolated habitats for differences based on guilds of dispersers. In the first, large and medium birds (except the fig-bird) with highly frugivorous diets declined in fragments, compared to continuous forest sites, whereas small-gaped birds with mixed diets increased in fragments (Moran *et al.* 2004). In the second, more primates were recorded visiting fruiting trees at mainland sites than at islands isolated for 10 years in French Guiana, but no differences in bird assemblages were detected between the two locations (Ratiarison & Forget 2005).

Interestingly, many studies on the effects of fragmentation on the seed dispersal of a single-species focus on endemics (Cordeiro & Howe 2003, Galetti *et al.* 2006, Ch. 2). For all of these studies, fragmentation (and sometimes hunting) reduces seed dispersal for both large (Cordeiro & Howe 2003, Ch. 2) and small-seeded (Galetti *et al.* 2006) endemic species. Therefore, it is possible that endemic species have more specialized dispersers and are especially susceptible to fragmentation, regardless of their seed size.

**Conclusions**

This study is significant because it compares forest fragmentation effects on seed dispersal for two coexisting species with contrasting dispersal agents, using direct counts of dispersed seeds to characterize seed dispersal in forest fragments and continuous forest. In forest fragments, small-seeded species like *B. multiflora* may continue to receive adequate seed dispersal needed to persist as healthy populations of seedlings and adults. The fate of large-seeded species such as *D. cestroides* seems to be quite the opposite. In forest fragments large-seeded species have reduced seed dispersal making them more susceptible to extirpation. Small-
seeded species are probably more resilient to fragmentation than large-seeded species because they have a greater number of generalist dispersers that are less prone to extinction and that utilize secondary forests in the surrounding matrix. In addition, endemic species may be more threatened by fragmentation regardless of seed size. Accordingly, conservation of large-seeded species and their dispersal agents will require reserves that are large enough to harbor reproductive populations of both mutualists.
CHAPTER 5

USE OF A RAPID SURVEY TO TEST FOR EFFECTS OF FOREST FRAGMENTATION ON FRUIT PRODUCTION: PRELIMINARY FINDINGS FROM FOUR SPECIES
Introduction

Fragmentation induces biotic changes to plant and animal communities and abiotic changes to the physical environment that can have severe impacts on plant populations. Responses of tree species and their animal mutualists to these effects of forest fragmentation vary (Ch. 2), so identifying the most threatened species is important for managing remaining tree populations in fragments. The high alpha diversity characteristic of tropical forests – the Central Amazon has over 1000 species of trees in 61 families and 288 genera (Gascon & Bierregaard 2001) – makes it difficult to devote in-depth studies to each species. Although generalizations based on species characteristics, i.e., seed dispersal of large-seeded species is more threatened than that of small-seeded species (Ch. 4), are valuable, they are not direct assessments of individual species. Therefore, surveys that can quickly and accurately identify threatened species will be useful in rapidly assessing the plant community.

This study has two goals. First is to examine how fragmentation affects fruit production, measured by fruit fall. Second is to test the effectiveness of a rapid survey used to sample fruit fall. Reduced fruit production in fragments has been attributed to the extirpation of animal pollinators (Dick 2001, Powell & Powell 1987), changes to pollinator behavior (Aizen & Feinsinger 1994), reduced outcrossing (Cascante et al. 2002), and resource limitation (Chacoff et al. 2004). In addition, disruptions of pollination mutualisms are hypothesized to pose the greatest risk for trees in fragments (Didham et al. 1996, Gilbert 1980, Howe 1977, Tabarelli et al. 2004) and may be an important indicator that a species is threatened by forest fragmentation. Here I present data comparing fruit fall of four tree species growing in continuous forest with those of conspecific individuals in forest fragments. I sampled fruit fall of three species using the rapid survey, and a fourth as part of a seed dispersal census.
Methods

Research was conducted in the experimentally fragmented forests and nearby continuous forest plots associated with the Biological Dynamics of Forest Fragment Project (BDFFP). Fragments there were isolated from large tracts of continuous forest 25 years ago and are periodically re-isolated from regenerating secondary forest by clearing a 100-m wide strip around each reserve. The project’s phytodemographic database was used to locate all individuals of identified fruiting species from within the 66-ha of mapped forest across all reserves.

In the wet season (January-March) of 2005, all fruiting tree species were censused. Fruit fall of species with at least two fruiting individuals in both fragments and continuous forest could be sampled using the rapid survey. Low fruiting overall in 2005 resulted in the identification of only three species that met this criteria; Manilkara bidentata (A. DC.) A. Chev., Ouratea sp. 1, and Micrandropsis scleroxylon (W.A. Rodrigues). Fruit fall of these three species were rapidly surveyed using either seed traps or wedge-shaped transects. Low replicates for each species resulted from the paucity of fruiting even within species in 2005 (Table 5.1).

Seed traps were used to survey fruit fall of M. bidentata (Sapotaceae) and Ouratea sp. 1 (Ochnaceae). Manilkara bidentata is a shade-tolerant evergreen that reaches the canopy (Crow 1980) and has fleshy red fruits. Ouratea sp. 1 produces oblong fruits that are black when ripe. The number of seed traps under the crown represented 3% of each individual’s total crown area. Crown area was estimated by measuring the distance from the crown center to crown edge at 20’ intervals and then summing the area of all 18 wedges. Single traps were also placed at 2, 5 and 10 m from the edge of the crown for two focal trees of each species. These traps were used to monitor the amount of fruit deposited at distances beyond the crown, which would estimate the
proportion of fruit missed by trapping exclusively under the crown. Traps remained in place until there was no fruit remaining on the parent tree (Table 5.1).

<table>
<thead>
<tr>
<th>Species</th>
<th>Sampling Method</th>
<th>Year sampled</th>
<th>Days sampled</th>
<th>Forest Size</th>
<th>Individuals Visited</th>
<th>Fruiting Individuals (surveyed)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>B. multiflora</em></td>
<td>Traps</td>
<td>2003</td>
<td>36.8 ± 2.8</td>
<td>1-ha</td>
<td>1 (100)</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>10-ha</td>
<td>3 (100)</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>100-ha</td>
<td>3 (100)</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>CF</td>
<td>5 (100)</td>
<td>4</td>
</tr>
<tr>
<td><em>M. bidentata</em></td>
<td>Traps</td>
<td>2005</td>
<td>46 ± 4.3</td>
<td>1-ha</td>
<td>5 (100)</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>10-ha</td>
<td>9 (35)</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>100-ha</td>
<td>16 (100)</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>CF</td>
<td>28 (58)</td>
<td>2</td>
</tr>
<tr>
<td><em>M. scleroxylon</em></td>
<td>Transect</td>
<td>2005</td>
<td>n/a</td>
<td>1-ha</td>
<td>16 (25)</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>10-ha</td>
<td>23 (92)</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>100-ha</td>
<td>84 (79)</td>
<td>3</td>
</tr>
<tr>
<td><em>Ouratea sp. 1</em></td>
<td>Traps</td>
<td>2005</td>
<td>67 ± 2</td>
<td>1-ha</td>
<td>1 (100)</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>10-ha</td>
<td>11 (100)</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>CF</td>
<td>11 (100)</td>
<td>2</td>
</tr>
</tbody>
</table>

Wedge-shaped transects were used to survey fruit fall of *Micrandropsis scleroxylon* (Euphorbiaceae). Fruits are dry, green, and segmented. A single wedge-shaped transect under each parent tree had an 80° base angle, and represented 22% of the total area beneath the crown. Near the end of the fruiting season a single count was made of all fruit, seeds, and fruit pieces from within these transects.

Fruits and seeds were also collected for *Bocageopsis multiflora* (Mart.) (Anonaceae), a canopy tree that produces small, fleshy orange to red fruits 0.8 cm in diameter. Seeds of this species were collected from seed traps as part of study on seed dispersal (Ch 4). Methods were
dissimilar to the rapid survey in that only a single trap was placed under the crown and nine other traps were placed at distances between 5-35 m from the edge of the crown to census seed dispersal.

All traps and transects were placed under areas where there was fruit in each individual’s crown. All fruit, fruit parts, and seeds in seed traps were collected and counted approximately every other week. Fruit pieces were classified by the fraction of fruit they represented and conservatively grouped to estimate whole fruit numbers. Stems not attached to fruit and seeds without fruit were also collected and counted. The number of stems and seeds that exceeded the sum of fruits counted was added to the sum of total fruits. Since *M. scleroxylon* fruits are explosively dispersed and can break into 6 segments, they were often found in small pieces that were fractions of the total fruit. These fruit pieces were summed to calculate the most conservative estimate of whole fruits they represented.

T-tests for each species were used to compare the number of fruits in fragments (all fragments pooled for *M. bidentata*) versus continuous forest or between fragment sizes (*M. scleroxylon* only). Log-transformed values of total fruits improved data normality and homoscedasticity, and so were used for all three rapid survey species. For *B. multiflora*, a regression was used to determine if the number of fruits was dependent on fragment size (1-ha, 10-ha, 100-ha, and continuous forest assigned a value of 1000-ha); fruit fall and fragment size were log-transformed to linearize the fits.

**Results**

None of the three species from the rapid survey showed significant differences in fruit fall between fragments and continuous forests (*M. bidentata*: $t=-0.86, P=0.18$; *M. scleroxylon*: $t=-1.92, P=0.45$ *Ouratea* sp. 1: $t=3, P=0.84$). The regression for the relationship between forest
size and fruit production for *B. multiflora* showed that as forest size decreased, so did fruit fall, although the trend was not significant ($R^2=0.41$, $P=0.09$, Figure 5.1).

**Figure 5.1** There is a positive relationship between fruit fall and forest size for *B. multiflora*, although it is not significant. LogSizes 0, 1, and 2 represent forest fragments of 1, 10, and 100-ha respectively while LogSize 3 represents continuous forest.

**Discussion**

The rapid survey failed to detect significant differences between fruit fall in fragments and continuous forest for all three species. The lack of significant effects could indicate either an absence of differences between fragments and continuous forest or a Type II error.

Supra-annual fruiting patterns and clumped distributions of rare tropical trees contribute to the difficulty of sampling using this rapid survey. The high frequency of supra-annual fruiting patterns among tropical trees results in highly variable fruit production within species between years and even between individuals within years (Camacho & Orozco 1998, Herrera *et al.* 1998). Given this intraspecific variability, data from only a few individuals for a single year may not be adequate to detect significant trends in fruit production between fragments and continuous forest.
However, because tropical trees occur at low densities (Hubbell & Foster 1986) obtaining large sample sizes is difficult. Upon isolation, a subset of tree species are completely eliminated from forest patches (Laurance et al. 2002). As a result, some species are represented by very few or no individuals across all fragments, making comparisons with individuals in continuous forest impossible. Here, despite extensive surveys within 66-ha of mapped forest at the BDFFP and over three months (Table 5.1), I only identified three species with at least two fruiting individuals in both fragments and continuous forest. Studies involving more than four individuals in each forest size and possibly several years of data are necessary to test for differences between fruit fall in fragments and continuous forest of a single species.

The only relationship between fruit fall and forest size that was close to significant was that of *B. multiflora*. As with *D. cestroides* (Ch. 3), reduced fruit fall with decreasing forest size for *B. multiflora* is likely a result of disruptions to pollinators or resource limitation in fragments. *B. multiflora* flowers are small (5-7 mm diameter), protogynous, have a diurnal antithesis, and a sweet but slightly rancid odor (Webber & Gottsberger 1995). Pollination is by thrips (order Thysanoptera), who were the only insects recorded visiting flowers (Webber & Gottsberger 1995) and may represent obligate pollinators for *B. multiflora*. The effect of fragmentation on thrips is unknown, but in fragments the most dramatic declines in pollination occur for rare and specialized pollinators, as they represent the most threatened mutualisms (Aizen & Feinsinger 1994, Renner 1998).

Censusing fruit production and seed dispersal using seed traps may not be appropriate for all species. For two of the three species, seed traps at three distances beyond the crown were used to account for dispersal beyond the crown, but no fruits, fruit parts, or seeds were captured in any of these traps. As a single trap only represents 0.5 m², it is likely that failure to capture
dispersed seeds results from an inadequate trapping area beyond the crown, rather than an absence of seed dispersal. Seeds beyond the crown were only found for *B. multiflora*, which had three times as many traps beyond the crown as any other species. Changes in rates of fruit removal in fragments or continuous forest would affect the number of fruits that fell underneath the crown. Therefore, the interpretation of the three species from the rapid survey is further complicated by unknown rates of removed fruit.

**Conclusions**

This study indicates that the rapid survey used here to assess fruiting of tropical trees in fragments and continuous forest is not adequate. Supra-annual fruiting, intraspecific variability, and rarity of tropical trees contribute to the difficulty of assessing many species within a short period of time. When more individuals were sampled, as for *D. cestroides* (Ch. 3) and *B. multiflora*, significant and nearly significant trends reveal lower fruit production with decreasing forest sizes. Studies involving fruit production of tropical trees need to sample more than 2-3 individuals in each treatment, or few individuals over several years. Research should target species of concern, like those with rare and specialized animal mutualists, in forest fragments.
CHAPTER 6

CONCLUSIONS
Tropical rainforests harbor a majority of the world’s biodiversity. Unfortunately, these forests are experiencing rapid deforestation and forest fragmentation. Fragmentation impacts species diversity and interactions within fragments, altering the species composition of remaining forest remnants. Although many single-species effects of forest fragmentation have been studied, few studies have examined the effects of fragmentation on multi-species interactions. In this dissertation I explored the effects of forest fragmentation on species interactions important for the regeneration of tropical rainforest trees – seed dispersal and seed predation – as well as seedling germination and establishment.

I demonstrated that fragmentation disrupts the seed dispersal mutualism for *Duckeodendron cestroides*. In wet tropical forests, close to 90% of tree species rely on animals for seed dispersal (Frankie *et al.* 1974, Howe & Smallwood 1982, McKey 1975, Webb & Peart 2001). As many animals are threatened by fragmentation, plants that depend on these animals for seed dispersal will experience changes to seed distributions. Adequate seed dispersal is the first step in recruitment for the majority of plants. Seed dispersal decreases potential competition with siblings and parents, increases the likelihood that seeds will escape density-dependent mortality (Connell 1971, Howe & Miriti 2000, Janzen 1970) and increases the probability of seeds arriving in favorable habitats (Augspurger 1984, Howe & Smallwood 1982). Here, forest fragmentation reduced the number and distance that seeds of *D. cestroides* were dispersed past the crown of the parent plant in two of three years. This discrepancy in seed dispersal in fragments and continuous forest resulted in fewer first-year seedlings in fragments, especially at distances close to the parent tree. Smaller populations of recruiting seedlings in forest fragments could contribute to *D. cestroides*’ decline there.
Much lower fruit fall was recorded for *D. cestroides* individuals in smaller forest sizes, 10 and 100-ha fragments, than in continuous forest. Fruit production fluctuated over the three years, being much lower in 2003 than in 2002 and 2004. Fruit production is dependent upon available resources and successful pollination. As *D. cestroides* flowers are pollinated by sphingid moths which often exhibit large home ranges, reduced pollinator movement could result in lower fruit fall in forest fragments. Alternatively, changes to the microclimate of fragments, such as decreased humidity and soil moisture, and increased temperatures (Kapos 1989, Laurance *et al.* 2002) can impose greater physical stress on tree species and limit resources for fruit production.

Post-dispersal seed fate of *D. cestroides* was also different in fragments and continuous forest. In continuous forest, seed removal by seed predators or secondary dispersers was high, over 75% of available seeds, but seed predation in fragments was only 6% of that in continuous forest. Just as dispersers of *D. cestroides* are reduced in forest fragments, animal seed predators also appear to be threatened. Reduced seed predation in fragments may also indicate reduced secondary dispersal and scatter-hoarding, and prove detrimental to seedling establishment.

Seedling germination was also different between fragments and continuous forest. Whereas seeds in continuous forest germinated within the same dry season as fruit fall, seeds in fragments delay germination until the rainy season when more moisture is available.

Despite changes to the processes leading up to successful seedling establishment (fruit production, seed dispersal, seed predation, and seed germination) I did not detect any differences in patterns of *D. cestroides* seedling establishment or mortality between fragments and continuous forest over three years. I expected that distance- and density-dependent mortality would influence the rates of seedling establishment and mortality more in fragments where
reduced seed dispersal results in seeds concentrated near the parent plant. Although distance-
and density-dependent morality did not affect the young seedlings studied here, changes in
seedling germination patterns and the number and distributions of first-year seedlings may affect
the long-term sustainability of *D. cestroides* seedlings and juveniles in forest fragments.

The effects of fragmentation observed for *D. cestroides*, may be especially harmful to rare
trees. Many tropical tree species occur at low densities (Hubbell & Foster 1986, Volkov *et al.*
2003). Fragmentation that reduces and isolates populations may cause extirpation of rare species
from fragments. For endemic trees that are globally rare, the disappearance of trees from
fragments can lead to extinction. Endemic species, like *D. cestroides*, may be at greater risk
from lost animal mutualists in fragments as they have more specialized species interactions.
Changes to fruit production, seed dispersal, seed predation, and germination could be the first
sign that populations of *D. cestroides* are in jeopardy.

None of the measures of seed dispersal for *Bocageopsis multiflora* – number, percentage,
distance, or distributions of dispersed seeds – were different in fragments or continuous forest.
In contrast to large-seeded species, like *D. cestroides*, small-seeded species, like *B. multiflora*,
are more resilient to fragmentation because they have more animals that can handle and disperse
their seeds and fewer dispersers that are sensitive to fragmentation. Therefore, forest
fragmentation may be more detrimental to species interactions for large-seeded species than
small-seeded species. Other fragmentation research supports this hypothesis with three lines of
evidence: 1) Large-seeded plant species are more prone to extinction, 2) Fragmentation restricts
or alters the movement of large animals more than small animals, and 3) Large and small-seeded
species seem to be differentially linked with primary and secondary forest habitats. In forest
fragments, large-seeded species should be a priority for conservation.
The rapid survey I used was not sufficient to examine the effect of forest fragmentation on fruit production. There were no significant effects of fragmentation on fruit fall of any of the four species surveyed. Supra-annual fruiting and rarity of tropical trees contributed to the identification of few fruiting individuals in fragments and continuous forest. Low replicate size, in combination with high intraspecific variation of fruiting individuals, likely caused an exaggerated Type II error rate. Rare tropical tree species need to be studied for several consecutive years in order to obtain enough data to draw conclusions on fruiting and reproduction.

Maintenance of species interactions within forest fragments is key to overall forest stability. The disintegration of a single relationship can be exponentially disruptive to plant and animal species directly and indirectly through complex relationships (Gascon et al. 2002). The conservation of forest fragments will depend upon the strength and sustainability of the plant community as well as its species interactions. Effects of disrupted species interactions on trees may be difficult to detect because the results take longer to surface in such long-lived species. Therefore, by studying the processes involved in plant regeneration, mutualisms (i.e., pollination and seed dispersal), and other interactions such as seed predation, changes to the next generation of plants can be detected years before they become apparent in the adult community. Reduced seed dispersal, seedling germination, or fruit set in fragments indicate that regeneration is in jeopardy. Because processes of dispersal limitation are important in maintaining diversity in tropical forests (Harms et al. 2000), altered regeneration processes of one species can change the dynamics of other species by changing seed and seedling communities. Without management for threatened species or threatened species interactions, species composition in fragments will be
heavily weighted towards resistant species and drastically altered from the original continuous forest.
LITERATURE CITED


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Oliveira, A. A. de 1997. Diversidade, estrutura e dinâmica do componente arbóreo de uma floresta de terra firme de Manaus, Amazonas. Instituto de Biociências da Universidade de São Paulo, São Paulo, Brazil.


APPENDIX: LETTERS OF PERMISSION

From: Jennifer Cramer  
To: jaboury.ghazoul@env.ethz.ch  
Date: Monday, March 5, 2007 10:52:00 AM  
Subject: Re: Biotropica - Decision on Manuscript ID BITR-06-310.R1

Dear editor,

I am writing to request permission to include the manuscript entitled "Forest fragmentation reduces seed dispersal of Dmecodendron cestroides, a Central Amazon endemic," manuscript ID BITR-06-310.R1, accepted for publication in Biotropica on December 28, 2006, as part of my doctoral dissertation from Louisiana State University.

Thank you,

Jennifer Cramer  
Ph.D. Candidate  
Department of Biological Sciences  
Louisiana State University  
Baton Rouge, LA 70820  
225-578-2459

From: Christopher Kaiser  
To: Jennifer Cramer  
Date: Tuesday, March 6, 2007 3:13:29 AM  
Subject: Re: Fw: Biotropica - Decision on Manuscript ID BITR-06-310.R1

Dear Jennifer,

Thank you for your enquiry. After acceptance, authors may use all or part of their article without modification in other publications of their own work. They may also use the article within their institution for educational purposes. This applies also to dissertations. So please feel free to include your accepted manuscript in your PhD dissertation.

Please do not hesitate to contact me should you have any further questions.

Best regards,

Christopher
From: Jennifer Cramer [mailto:jencramer@yahoo.com]
Sent: 28 March 2007 18:16
To: Stone, Sharon (ELS-EXE)
Subject: Re: Your paper [BIOC_3379] submitted to Biological Conservation

Sharon Stone,
I hope that you can help me with a problem I am having getting approval to use the manuscript as part of my dissertation. I have submitted requests directly to the Global Rights Department (March 5) and also using the online support and contact (March 20) but have not heard back from either of these requests. I need written permission from Elsevier to publish the article "Forest fragmentation differentially affects seed dispersal of large and small-seeded tropical trees" (BIOC_3379) as part of my dissertation. I also need this permission within the next week so that I can submit my dissertation by the deadline for the graduate school here at my university. I realize that this is not necessarily your department, but felt it necessary to email a person directly as my previous emails have gone unheaded. I hope that you can forward this email to someone to help me get my letter of permission.
Thank you for your time,
Jen Cramer

Jennifer Cramer
Ph.D. Candidate
Department of Biological Sciences
Louisiana State University
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225-578-2459
2 April 2007

Ms Jennifer Cramer
Louisiana State University
Jcrame5@lsu.edu

Dear Ms Cramer

BIOLOGICAL CONSERVATION, Accepted for publication (BIOC 3379), Cramer, “Forest fragmentation...”

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Jennifer Mei Cramer was born in Honolulu, Hawaii, on March 21, 1976, to Roger and Katherine Cramer. She grew up in Honolulu where she lived until she graduated from Kalani High School in 1994 as a valedictorian. Jennifer began Earlham College in Richmond, Indiana, in the fall of that same year. Earlham’s strong biology curriculum, with an emphasis on ecology, led Jennifer to declare a degree in biology. A student research project on the Fruit Flag Hypothesis inspired Jennifer’s pursuit of a career in ecological research, specifically mutualistic processes such as seed dispersal. In June 1998 Jennifer graduated from Earlham with a Bachelor of Arts in biology. In 1999 Jennifer began pursuing her doctorate in the Department of Biological Sciences at Louisiana State University. Under the guidance of Dr. G. Bruce Williamson, Jennifer traveled to the Biological Dynamics of Forest Fragments Project in Brazil for her field research on forest fragmentation effects on mutualisms and regeneration of *D. cestroides*. She was named a 2007 Presidential Management Fellow and will be starting a position at the Santa Fe National Forest after graduation.