

# Habitat use and seed removal by invasive rats (*Rattus rattus*) in disturbed and undisturbed rain forest, Puerto Rico

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## Abstract

Despite frequent occurrences of invasive rats (*Rattus* spp.) on islands, their known effects on forests are limited. Where invasive rats have been studied, they generally have significant negative impacts on native plants, birds, and other animals. This study aimed to determine invasive rat distribution and effects on native plant populations via short-term seed removal trials in tropical rain forest habitats in the Luquillo Experimental Forest, Puerto Rico. To address the first objective, we used tracking tunnels (inked and baited cards inside tunnels enabling animal visitors' footprints to be identified) placed on the ground and in the lower canopy within disturbed (treefall gaps, hurricane plots, stream edges) and undisturbed (continuous forest) habitats. We found that rats are present in all habitats tested. Secondly, we compared seed removal of four native tree species (*Guarea guidonia*, *Buchenavia capitata*, *Tetragastris balsamifera*, and *Prestoea acuminata*) between vertebrate-excluded and free-access treatments in the same disturbed and undisturbed habitats. Trail cameras were used to identify animals responsible for seed contact and removal. Black rats (*Rattus rattus*) were responsible for 65.1% of the interactions with seeds, of which 28.6% were confirmed seed removals. Two plant species had significantly more seeds removed in disturbed (gaps) than undisturbed forest. *Prestoea acuminata* had the lowest seed removal (9% in 10 days), whereas all other species had >30% removal. Black rats are likely influencing fates of seeds on the forest floor, and possibly forest community composition, through dispersal or predation. Further understanding of rat-plant interactions may be useful for formulating conservation strategies.

Abstract in Spanish is available with online material.

## KEYWORDS

forest gaps, ground and understory foraging, invasive species, Luquillo Experimental Forest, montane forest, native tree vulnerability, pre-hurricane frugivory, rodent seed predation

## 1 | INTRODUCTION

Invasive rats (*Rattus* spp.) are widely known for threatening human health by disease transfer and impacting economies by causing agricultural and horticultural damage (Pimentel, Lach, Zuniga & Morrison, 2000). Less is known about the effects of invasive rats on native habitats that are part of non-anthropogenic ecosystems,

yet several regional reviews have summarized such effects occurring over the last four decades (Banks & Hughes, 2012; Harper & Bunbury, 2015; Shiels, Pitt, Sugihara & Witmer, 2014). The global effects of invasive rats on island flora and fauna were extensively reviewed by Towns, Atkinson and Daugherty (2006). Their review described how these rats could be threatening global biodiversity in some insular ecosystems by causing species loss, whereas others

received less rat damage and greater native species persistence. Invasive rats can affect terrestrial ecosystem structure and function via frequent seed predation (Shiels & Drake, 2015) and interfere with ecological interactions that have coevolved throughout time (Traveset & Richardson, 2006) such as pollination and seed dispersal mutualisms (Drake & Hunt, 2009; Shiels et al., 2014). Rats have been linked with decline and extinctions of native vertebrate species globally (Towns et al., 2006), especially those species native to islands (Harper & Bunbury, 2015; Traveset & Richardson, 2006). A recent study provides evidence of how invasive rat presence on islands can alter both terrestrial fauna and marine ecosystem productivity and function (Graham et al., 2018). The black rat (*Rattus rattus*) has already reached and successfully colonized most of the world's islands, primarily as stowaways on ships (Atkinson, 1985; Drake & Hunt, 2009; Towns et al., 2006); it has been rated as the most damaging of all invasive rodents worldwide (Capizzi, Bertolino & Mortelliti, 2014), and especially so to island forests (Shiels et al., 2014).

The effects of rat invaders on island forests are often amplified relative to continental forests due to the few species, or absence, of native rodent species in most island ecosystems (Drake & Hunt, 2009). The challenge in determining whether invasive rats are altering native forest composition and structure is that rats target seeds for consumption and therefore their effects on forest stands may not be noticeable for 10s or 100s of years (Shiels & Drake, 2015; Shiels et al., 2014). Consequently, focused measurements on rat interactions with seeds and seedling recruitment have become standard methods to understand how invasive rats are affecting forest tree communities. On Palmyra Atoll in the tropical Pacific, black rat eradication resulted in an increase of the native seedling layer for five of six species studied, but also increased seedlings of a non-native palm (*Cocos nucifera*) 13-fold (Wolf et al., 2018). Not all plant species are equally vulnerable to black rat predation, as Shiels and Drake (2011) experimentally determined by examining 12 woody species in a Hawaiian forest; some species were highly vulnerable to rats (i.e., eight species with 50–100% fruit and seed removal, and six species with 60–100% seed predation) while others experienced no removal or predation.

In addition to rat preference for particular plant species, the habitats where such food resources are found may affect their foraging behavior (Law & Dickman, 1998) and perhaps the vulnerability of plant species to rat predation (Cox, Dickman & Cox, 2000). Disturbance creates variation in habitat, and many animal species prefer or avoid disturbed habitats based on the states of vegetation cover, prey, and predators (Law & Dickman, 1998). In Australia, Cox et al. (2000) found that black rats utilized forest more than open or scrub-dominated habitats, and their microsite preference was areas of deep leaf litter and dense understories with numerous vertical stems. In Hawaiian forests, invasive rats and house mice (*Mus musculus*) were under 30-cm-tall vegetation cover at least 88% of the time when they were out of their dens (Shiels, 2010). Additionally, black rats are avid climbers and use both the ground and canopy for denning and foraging (Dowding & Murphy, 1994; Shiels, 2010).

Despite black rats denning in tree cavities above the ground in New Zealand forest, they spent ca. 90% of the night active on the ground (Dowding & Murphy, 1994).

Little is known about invasive rat ecology, habitat use, and impacts to our tropical study site in the Luquillo Experimental Forest (LEF) of Puerto Rico (Willig & Gannon, 1996). Invasive rats in the LEF have been recorded consuming litter and arboreal invertebrates, dead wood, snails, and some plant parts, including fruit and seed (Willig & Gannon, 1996). Weinbren, Weinbren, Jackson and Villella (1970) conducted caged no-choice feeding trials with fruits of 11 tree species in the LEF by offering them to four individual black rats. They found that rats ate portions of all offered fruit, but some species were eaten more than others. Black rats are the only rat species known to be at our study site in the LEF based on past studies (Shiels & Ramírez de Arellano, 2018; Weinbren et al., 1970; Willig & Gannon, 1996) and evidence that the few native rodents in Puerto Rico went extinct shortly after European colonization (Turvey, Oliver, Nargane Storde & Rye, 2007). The Norway rat (*R. norvegicus*) is established in Puerto Rico, but this rat is typically found in urban settings or in active or abandoned agricultural fields rather than in forests (Shiels, 2010; Shiels et al., 2014).

Our present study in the LEF first aimed to determine habitat use and distribution of invasive rats in disturbed and undisturbed sites, including whether they occur in the lower canopy and on the ground, and secondly, to assess short-term fruit and seed removal of four native tree species by rats. We sought answers to the following questions: (1) Are rats more likely to be found in disturbed (forest canopy gaps) than undisturbed habitat? (2) Are rats more likely to be active on the ground or in the lower canopy? (3) Which fruits and seeds from the LEF are rats attracted to, and which are removed, at the forest floor? (4) How does fruit and seed removal of particular tree species by rats compare to those of other vertebrates in the same forest? We expected rats would be more common in the lower canopy than on the ground because of their arboreal habits, and within disturbed habitats rather than undisturbed due to their affinity for more dense vegetation cover near the ground that develops under canopy gaps. Additionally, we expected rats to remove most of the native fruits and seeds offered to them on the ground because of their opportunistic foraging behavior.

## 2 | METHODS

### 2.1 | Study site

The study was conducted in the LEF in northeastern Puerto Rico, specifically at the northwest section of the forest near El Verde Field Station (EVFS; 18°20' N, 65°49' W), at 300–485 m of elevation. The LEF, a 19,650-ha tropical evergreen forest, is an insular ecosystem not only because it is on an island, but also because the forest itself is surrounded by an urban and suburban landscape. Mean annual precipitation at EVFS is 346 cm, and there is high year-to-year variation and mild seasonality; mean monthly temperature is 25°C (Brokaw et al., 2012). The average canopy height near EVFS is  $18.1 \pm 0.3$  m

(Shiels & González, 2014). Disturbances in this forest, such as landslides, hurricanes, treefalls, and floods, result in distinct habitats that affect resource heterogeneity and redistribution of animal populations (Reagan & Waide, 1996). There have been three major hurricanes (i.e., Categories 3–5 on Saffir–Simpson scale) that have passed over the LEF in the last 30 years. Our study was completed 2 months prior to Hurricane Maria passing over the LEF on 20 September 2017.

The sites used to determine the effect of closed canopy forest versus canopy gaps on rat presence and activity were as follows: (a) closed canopy forest ( $N = 10$ ), which had no significant canopy disturbance (from experiments, hurricanes, or treefalls) for the previous 19 yrs, and each plot was at least 30 m from any gap or stream, (b) stream gap ( $N = 8$ ), which was defined as 1–4 m from the stream edge (determined by the recent high water line and presence of bare rocks), and (c) forest gap ( $N = 8$ ), which included a gap  $8 \times 12$  m created by a recent (<1 year) treefall, a gap  $8 \times 20$  m created by trimming the canopy around a powerline (ca. 3 years prior), and six  $30 \times 30$  m gaps ( $N = 3$  that were 2.5 years old,  $N = 3$  that were 12 years old) created to simulate hurricane damage within the canopy trimming experiment (CTE; Shiels & González, 2014) (Figure 1). Each of the three CTE blocks (A, B, and C) contain four  $30 \text{ m} \times 30 \text{ m}$  plots, where a smaller  $20 \text{ m} \times 20 \text{ m}$  core area was used for data collection and measurement. The CTE forest gap plots used in our study had all the trees >15 cm dbh trimmed of all branches <10 cm diameter, all palm fronds trimmed while preserving the apical meristem, and all trees 10–15 cm dbh cut (topped) at 3 m height (Richardson, Richardson, González, Shiels & Srivastava, 2010; Shiels et al., 2010). The trimming of the CTE plots was first completed in November 2004–June 2005, and since then, three plots were re-trimmed in November 2014. The vegetation status, canopy openness, and details of each treatment within the CTE, including the closed canopy (control) plots, are

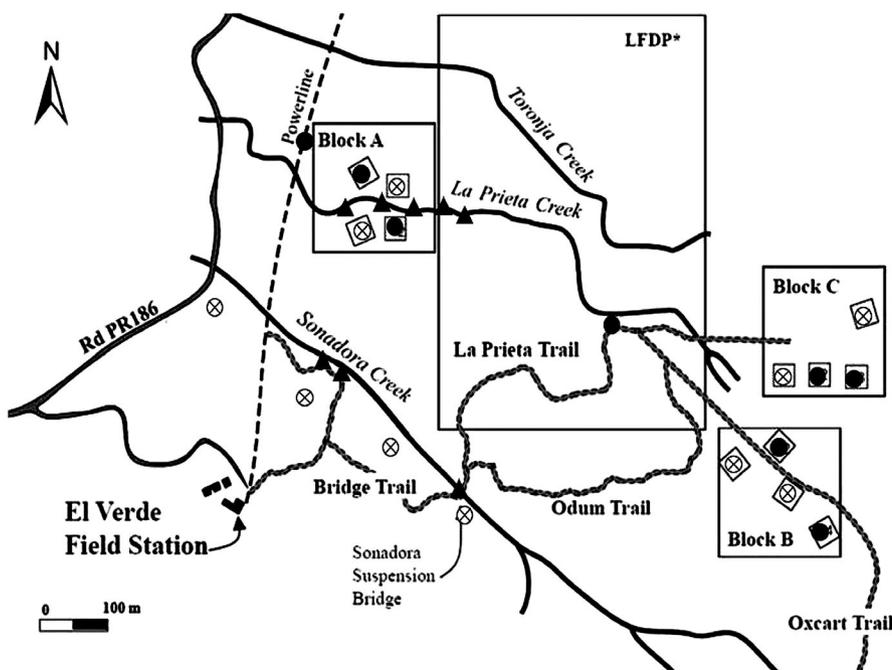
summarized in Shiels et al. (2010) and Shiels and González (2014). The forest gaps we used in our study (0.01–0.09 ha) were similar in size as most gaps created by wind storms in this section of the LEF (Brokaw & Grear, 1991; Shiels & González, 2014).

## 2.2 | Tracking tunnels

To assess rat activity and habitat use in gap and closed canopy forest habitats, we used tracking tunnels, which are inked and baited cards placed in tunnels enabling animal visitors' footprints to be identified (Shiels, 2010; Shiels & Ramírez de Arellano, 2018). In 23 representative plots (out of the 26 used in the latter part of the study; Figure 1), two tracking tunnels that were 60 cm long with  $10 \times 10$  cm openings were randomly placed approximately 10 m apart from each other (near the gap center in forest gaps); one tunnel was placed on the ground, and the other tunnel was secured by plastic straps on a branch or liana in the lower canopy at an average height of 143.5 cm (SE = 7.6 cm). Paper cards placed in each tracking tunnel had bait in the middle, which was surrounded by ink. The bait used was a  $2 \times 2$  cm coconut chunk placed on top of Skippy peanut butter. All tracking tunnels, cards, and ink were purchased from Pest Control Research LP ([www.traps.co.nz](http://www.traps.co.nz)). To represent each of the different habitats in which tunnels were placed, we chose seven stream gap replicates ( $N = 7$ ), seven forest gaps ( $N = 7$ ), and nine plots under closed canopy forest ( $N = 9$ ). A total of 46 tunnels were placed accordingly across the 23 chosen plots on 4 June 2017 and checked for footprints 1 day and 2 days later.

## 2.3 | Seed removal trials

Trials were also conducted to determine whether rats removed native seeds from the forest floor. The four tree species (Table 1) were



**FIGURE 1** A map of the El Verde forest within the Luquillo Experimental Forest, Puerto Rico, showing the 26 plots used in this study. The circles with crosses represent closed canopy forest, the solid circles indicate forest gaps, and the solid triangles are stream gaps. Blocks A, B, and C belong to the canopy trimming experiment. Map modified from Richardson et al. (2010)

**TABLE 1** Seed characteristics of the four species of native trees used in the field trials in the Luquillo Experimental Forest, Puerto Rico. For seed measurements,  $N = 6\text{--}30$  seeds for each species, and values are displayed as mean  $\pm$  SE

Species	Family	Length (mm)	Width (mm)	Mass (mg)
<i>Guarea guidonia</i> ( $N = 28$ )	Meliaceae	11.6 $\pm$ 0.34	8.29 $\pm$ 0.13	4.27 $\pm$ 0.19
<i>Buchenavia capitata</i> ( $N = 6$ )	Combretaceae	20.5 $\pm$ 1.12	11.65 $\pm$ 1.25	91.67 $\pm$ 0.83
<i>Tetragastris balsamifera</i> ( $N = 13$ )	Burseraceae	19.16 $\pm$ 0.92	16.29 $\pm$ 0.50	14.10 $\pm$ 1.92
<i>Prestoea acuminata</i> ( $N = 30$ )	Areaceae	13.56 $\pm$ 0.19	13.14 $\pm$ 0.66	12.44 $\pm$ 0.47

chosen based on their fruit and seed availability during the trial period (June–July 2017; all species trialed were fruiting during this period). *Prestoea acuminata* and *Tetragastris balsamifera* are both within the top 15 most common woody species in the forest, whereas *Guarea guidonia* and *Buchenavia capitata* are slightly less abundant but are often larger canopy species (Shiels et al., 2010). Plant species are hereafter referred to by genus. All fruits and seeds were collected freshly fallen from the forest floor near EVFS and stored in a refrigerator to slow their decomposition until used in the trials. *Buchenavia* fruit was collected beneath two trees, and only fruit with yellow coloring (its natural color when ripe) and < 10% damage from invertebrates were used in trials. *Guarea* seeds were collected from seven individuals, and *Tetragastris* seeds were collected from two individuals. *Guarea* and *Tetragastris* seeds were trialed without their fruit because they have dehiscent fruit and generally shed their seeds in the canopy with the fruit covering maintained on the tree. All aforementioned fruit or seeds (hereafter seeds) were collected 1–10 June 2017. *Prestoea* (sierra palm) seeds were collected below five individuals on 16 July 2017. Some *Prestoea* included their natural thin fruit covering, while others did not; however, all seeds were freshly fallen and did not have evidence of beetle colonization (Janzen, 1972). Prior to placing the seeds in field plots, conspecific seeds were mixed and then chosen randomly to appear in each treatment station. The 26 plots shown in Figure 1 were all at least 20 m away from each other and each plot had two different treatments (totaling 52 stations), each 10 m apart within a plot. The two treatments per plot were randomly assigned as either: (1) Caged treatment, where the seeds were enclosed within a metal-mesh (1 cm aperture) cage (30  $\times$  30  $\times$  30 cm; lacking a floor) to exclude all vertebrates from accessing them, or (2) Open treatment, where seeds were placed on the forest floor without any caging, allowing all animals to freely access them. The number of seeds placed in each station was consistent per species yet varied from one to six among species: *Guarea* (6), *Buchenavia* (1), *Tetragastris* (2), and *Prestoea* (3). At 20 (seven closed canopy, seven forest gap, six stream gap) of the 26 Open treatment replicates, Reconyx HyperFire trail cameras (models HC500 and HC600) were positioned no more than 50 cm from the seeds, secured to a tree at 30–50 cm aboveground, and adjusted to face the seeds to monitor the organisms responsible for seed contact (see below) and removal. The camera settings varied slightly, but monitored continuously, and were set to take from 2 to 5 pictures 1 s apart every time they were triggered by motion. Some of the cameras were additionally set to take a picture at 1-hr intervals, which was helpful for filling information gaps (i.e., insect contact did

not trigger the cameras). The number of seeds removed from each station was monitored on 1, 3, 7, and 10 days after being set out. If seeds were missing on these days, then the Secure Digital (SD) card was replaced with a blank one and the pictures were observed for the following: date, time, animal pictured, contact with seed (including contact with any part of the animal's body, as well as consumption or removal), and removal of seed. At the end of each trial, and to ensure independence among trials, every treatment station (both Open and Caged) was moved at least 3 m to another random location in the plot. Trial dates in 2017 for each species were as follows: *Guarea* 11–21 June; *Buchenavia* 22 June to 3 July; *Tetragastris* 3–14 July; *Prestoea* 17–28 July.

## 2.4 | Statistical analysis

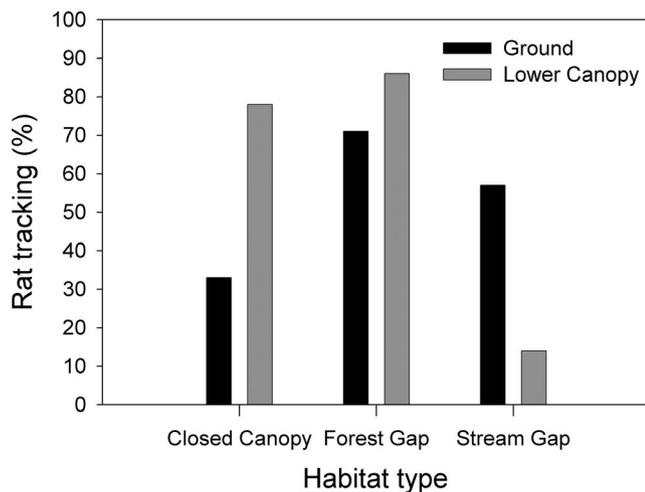
Rat activity (i.e., presence of rat tracks inside tracking tunnels after 2 days of observation) was examined for differences among the three habitats using separate chi-square tests for ground, lower canopy, and ground + lower canopy. Additional chi-square testing compared rat tracking on the ground versus lower canopy for each of the three habitat types. For seed removal trials at day 10, we used generalized linear models (GLMs) with either binomial errors (i.e., logistic regression to predict all seeds present vs. a seed removed) or Poisson errors (i.e., Poisson regression to predict counts of seeds removed in a plot). To assess evidence of any seed removal between Caged and Open treatments, we included all four species ( $N = 104$  plots per treatment) in GLM with binomial errors; once we determined that the Open treatments were indeed receiving frequent seed removal and Caged treatments were not ( $z = 5.090$ ;  $SE = 0.548$ ;  $p < 0.0001$ ; only 5 out of 312 seeds disappeared from Caged treatments during all trials), we excluded the Caged treatment for all subsequent analyses. We tested the effect of habitat type on seed removal at the end of the trial period (10 days) when all four species were included (binomial errors), and when each species was considered individually (Poisson errors for *Guarea*, *Tetragastris*, *Prestoea*; binomial errors for *Buchenavia*). All statistical analyses were conducted in R version 3.0.3, and significant differences were based on  $p < 0.05$ .

## 3 | RESULTS

There was no significant difference in rat tracking on the ground among habitats ( $\chi^2 = 2.390$ ;  $df = 2$ ;  $p = 0.3028$ ). However, there was

a significant difference in rat tracking in the lower canopy among habitats ( $\chi^2 = 9.272$ ;  $df = 2$ ;  $p = 0.0097$ ; Figure 2), as the stream gap had less rat tracking in the lower canopy (14.3%) than the closed canopy forest (77.8%) and the forest gap (85.7%; Figure 2). When ground and lower canopy tracking tunnel activity were combined for each plot, there was no significant difference among habitats ( $\chi^2 = 5.243$ ;  $df = 2$ ;  $p = 0.0727$ ), yet the stream gap tended to have the lowest rat tracking (35.7%) relative to the closed canopy forest (55.5%) and the forest gap (78.6%) (Figure 2). There were no differences in rat tracking between ground and lower canopy for any of the habitats compared (closed canopy:  $p = 0.1547$ ; forest gap:  $p > 0.9999$ ; stream gap:  $p = 0.2646$ ). Although rats made up the vast majority of the animal tracking, most of the tracking tunnels also had tracks of additional species, including 12 occasions of lizards, seven of coqui frogs (*Eleutherodactylus coqui*), 20 of snails, and 37 of insects.

Numbers of vertebrate-accessible seeds removed from the forest floor in each 10-day trial were 7–48 seeds, or 9.0% (*Prestoea*) to >30% (*Guarea* = 30.8%, *Buchenavia* = 34.6%, *Tetragastris* = 36.5%) of seeds trialed. When seed removal at 10 days for all four native tree species was combined, disturbed habitats (i.e., stream and forest gaps) tended to have more plots with at least one seed removed than those in closed canopy forest (Figure 3); however, these comparisons were not statistically significant (closed canopy vs. stream gap or forest gap:  $z = 1.686$ ;  $SE = 0.501$ ;  $p = 0.0919$ ). When seed removal among habitats was compared by species, *Guarea* had significantly more seeds removed in forest gaps than closed canopy forest ( $z = 2.106$ ;  $SE = 0.329$ ;  $p = 0.0352$ ) and stream gaps ( $z = 2.509$ ;  $SE = 0.391$ ;  $p = 0.0121$ ), but there was no difference in removal between closed canopy and stream gaps ( $p = 0.4951$ ; Figure 4). *Buchenavia* had significantly more seeds removed from stream gaps than closed canopy forest ( $z = 2.112$ ;  $SE = 1.282$ ;  $p = 0.0347$ ), whereas other comparisons were not statistically different (closed canopy vs. forest gaps:  $p = 0.1885$ ; forest gaps vs. stream gaps:



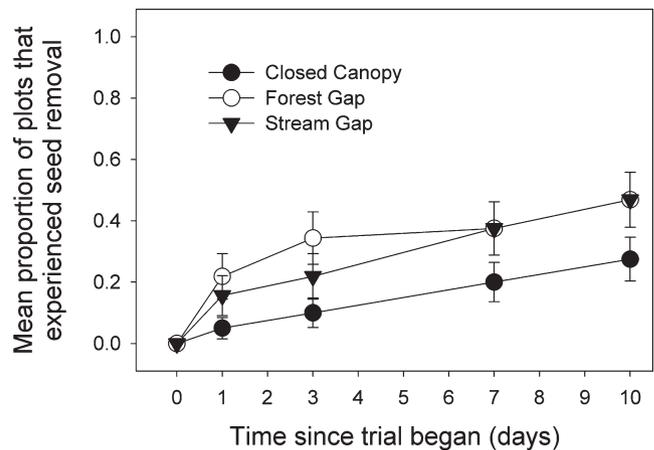
**FIGURE 2** Percentage of tracking tunnels that were tracked by rats on the ground ( $N = 23$ ) and in the lower canopy ( $N = 23$ ), in the three dominant habitat types (closed canopy, forest gap, and stream gap), in the Luquillo Experimental Forest, Puerto Rico

$p = 0.3230$ ; Figure 4). Although *Tetragastris* seed removal tended to be higher in stream gaps than other habitat types, this pattern was not significant ( $p = 0.2330$ ), nor was the closed canopy vs. forest gap ( $p = 0.7780$ ) and forest gap vs. stream gap ( $p = 0.1770$ ; Figure 4). Seed removal of *Prestoea* was the lowest of all tested species, and there were no significant differences when habitats were compared (closed canopy vs. forest gap:  $p = 0.1500$ ; closed canopy vs. stream gap:  $p = 0.4544$ ; forest gap vs. stream gap:  $p = 0.4320$ ; Figure 4).

Trail cameras recorded animal activity in the vicinity of the seed plots for the duration of the trials. Of the 86 occasions when an animal contacted a seed of the target species, 56 (or 65.1%) of those were black rats (Figure 5, Supporting Information Figure S1). Sixteen occasions (or 28.6% of rat contacts) were confirmed seed removals by black rats, and two occasions were removals by the ruddy quail dove (*Geotrygon montana*). Aside from the black rat, no other rodent species were pictured during our 40 days of continuously monitoring seeds in this forest. *Buchenavia* seeds had the most occasions of animal contact (41), while the remaining species had fewer: 17 for *Guarea*, 11 for *Tetragastris*, and 17 for *Prestoea* (Figure 5). In addition to those animals mentioned above and in Figure 5, the following were pictured near, but not in contact with, the seeds: feral cats (*Felis catus*; 10 pictures), mongoose (*Herpestes auropunctatus*; three pictures), and pearly eyed thrasher (*Margarops fuscatus*; two pictures).

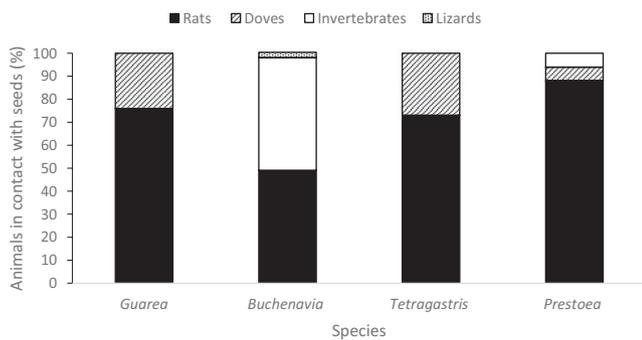
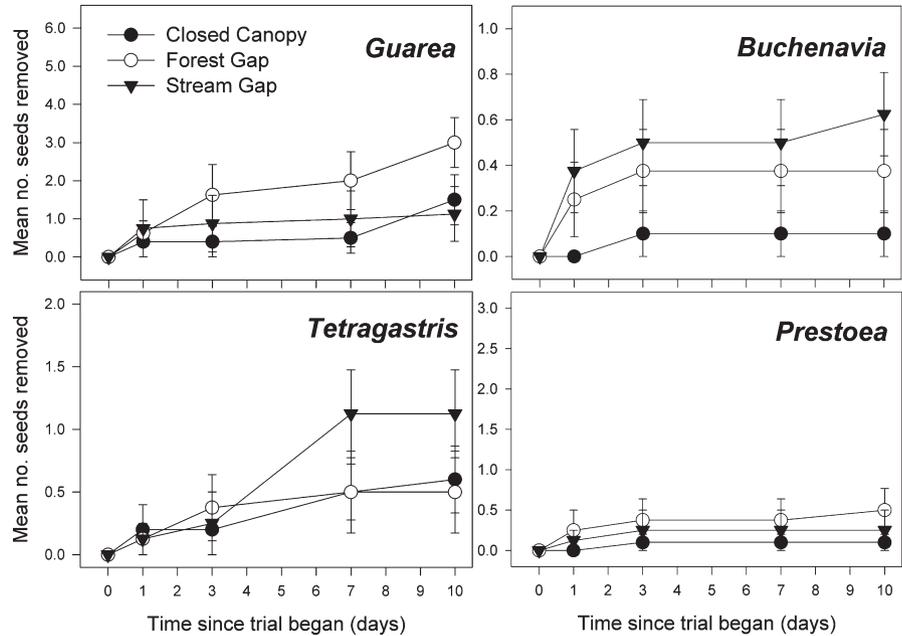
## 4 | DISCUSSION

Tracking tunnels and monitoring of seed removal trials showed rat presence on the ground and lower canopy in disturbed (forest and stream gaps) and undisturbed (closed canopy) patches of the LEF.



**FIGURE 3** Mean (SE) proportion of plots that had at least one seed removed from Open treatments in each of the three habitat types (closed canopy, forest gap, and stream gap) when all four tree species (*Guarea*, *Buchenavia*, *Tetragastris*, and *Prestoea*) were combined during the trial period of 11 June to 28 July 2017 in the Luquillo Experimental Forest, Puerto Rico. Seed removal tended to be greater in the disturbed habitats (forest gap and stream gap) than in the closed canopy forest, but there was no statistical difference among habitat types ( $p = 0.0919$ )

**FIGURE 4** Mean (SE) number of seeds removed in the Open treatment for each of the four species trialed in each of the three habitat types. At the beginning of each trial, the number of seeds per station varied by species: *Guarea* (6), *Buchenavia* (1), *Tetragastris* (2), and *Prestoea* (3). There were significantly more *Guarea* seeds removed from the forest gap habitat relative to the closed canopy ( $p = 0.0352$ ) and stream gap ( $p = 0.0121$ ); the stream gap habitat had more *Buchenavia* seed removal than the closed canopy ( $p = 0.0347$ )



**FIGURE 5** Percentage of images from trail cameras where animals contacted seeds in the Luquillo Experimental Forest, Puerto Rico. Contact was defined as an animal touching the seed with any part of its body, and this included seed consumption and removal. *Guarea* ( $N = 17$ ), *Buchenavia* ( $N = 41$ ), *Tetragastris* ( $N = 11$ ), *Prestoea* ( $N = 17$ )

Trail cameras used in our study, and evidence from past studies (Shiels & Ramírez de Arellano, 2018; Weinbren et al., 1970; Willig & Gannon, 1996), indicate that the most abundant, and probably the only, rat species in this forest is the black rat (*Rattus rattus*). Given the known omnivorous diets of black rats (Shiels et al., 2013, 2014), our findings of rat presence on the ground and aboveground in all major habitats highlight how invasive rats may interact with most terrestrial species and possibly some stream biota in this forest. Our seed removal trials showed that all four tree species tested are attractive and have seeds removed by black rats. *Guarea* and *Buchenavia* experienced greater seed removal in gaps than in closed canopy forest, and black rats appear to be minor threats to the sierra palm (*Prestoea*) because palm seeds were rarely removed from the forest floor. Black rat interactions with seeds could ultimately be affecting plant composition and structure in forests such as the LEF.

Wind and water disturbances in the LEF create forest gaps and a patchwork of habitats of different successional stages (Brokaw et al., 2012) that are expected to influence animal presence and behavior (Reagan & Waide, 1996). Forest gaps in the LEF are relatively small (0.01–0.09 ha; Brokaw & Gear, 1991; Shiels & González, 2014) and are a range of ages since prior disturbance (1–19 years in our study). Black rats were present in all of the forest habitats sampled (closed canopy, treefall gaps, hurricane plots, and stream edges), and on the ground and in the lower canopy. In Australia, Bennett (1990) found that black rats were more common in disturbed and fragmented forest than in continuous forest stands. Black rats are known to be the most arboreal of the invasive rodent species (Shiels et al., 2014), and they den and are active in the canopy and on the ground (Dowding & Murphy, 1994; Shiels, 2010). Although not statistically different, there was a tendency for greater rat activity in the lower canopy than on the ground in our closed canopy forest plots; escaping predators by utilizing aboveground habitat may be one possible explanation for such a trend, and feral cats and mongoose are known rat predators that were observed in our study plots. As a result of the increase in sunlight to the forest floor following canopy disturbance, the forest and stream gaps typically have thicker vegetation ground cover than does the closed canopy forest (Shiels et al., 2010). The thicker understory in the sampled gaps may explain why rat activity was equivalent between ground and lower canopy in the forest gaps, and significantly higher on the ground than in the lower canopy in the stream gaps. Black rats prefer habitats with dense understory cover (Cox et al., 2000; Shiels, 2010), and it may be more difficult for nocturnal predators to detect and catch rats in dense ground cover. Rats also may be more active on the ground in riparian areas due to the availability of free water and aquatic prey (Reagan & Waide, 1996; Willig & Gannon, 1996).

Prominent rat activity on the ground in forest gaps and stream gaps may have influenced the greater numbers of seeds removed

in these disturbed habitats for two of the four trialed tree species (*Guarea* and *Buchenavia*, respectively) relative to the closed canopy forest. In fact, *Guarea* had the most seeds removed of any species tested within forest gaps, which was the habitat with the highest rat tracking (78.6%). Fleshy fruit coverings can be attractive for consumption by black rats (Shiels & Drake, 2011), and fruit may comprise 55% of the black rat diet (Shiels et al., 2013). *Buchenavia* was the only species used in our trials that contained a thick and fleshy fruit covering, and previous LEF studies documented black rats consuming *Buchenavia* fruit (Weinbren et al., 1970) and seed (Willig & Gannon, 1996). Neither *Guarea* nor *Tetragastris* had fruit coverings in our trials but they had equivalent proportions of seeds removed as *Buchenavia*. Based on previous documentation, and our findings of initially high seed removal rates the first 3 days of trial and 41 incidents of animal contact recorded by the cameras over 10 days, *Buchenavia* is attractive to both black rats and invertebrates, and less so to lizards. Despite contact and some possible fruit consumption of *Buchenavia* by invertebrates (mainly insects) and lizards, only rats and ruddy quail doves (on two occasions) were documented removing the seeds of any trialed species. Although black rats removed seeds 28.6% of the incidences when pictured in contact with them, there were many occasions where the seeds were outside of the camera's view while black rats were pictured (two times with *Prestoea*, five with *Guarea*, seven with *Tetragastris*, and 22 with *Buchenavia*); these potentially missed rat-seed interactions almost certainly resulted in our underestimate of black rat contact and removal of seeds.

*Prestoea* palm had the fewest seeds removed (9%) at the end of the 10-day trial, despite frequent visits and occasional contact with the seeds by black rats (Figure 5). This finding disagrees with Snyder, Wiley and Kepler (1987), who suggested that this palm might be an important food source for the black rat, and also differs from earlier findings in Hawaii where endemic palms in the genus *Pritchardia* (Shiels & Drake, 2015), and Lord Howe Island where the endemic *Lepidorrhachis* palm (Auld, Hutton, Ooi & Denham, 2010), were highly susceptible to black rat removal and seed predation. Portions of offered *Prestoea* fruit were previously reported as consumed by black rats (Weinbren et al., 1970; Willig & Gannon, 1996), but evidence from our study demonstrates that the seeds are not favored by these rats during the summer season. *Prestoea* is known to be reproductively active throughout the entire year and produce hundreds of seeds per individual during each fruiting, but nearly all of the seeds produced (up to 99.7%) are eventually attacked and often depredated by a scolytid beetle (Janzen, 1972). Therefore, rather than invasive rats, the primary seed predator of *Prestoea* palm seeds is probably the native scolytid beetle.

Many factors can affect the likelihood of seed removal in the field, including habitat disturbance, animal densities, and available foods; yet documenting the frequency of seed removal and the animals involved are important first steps to determine seed fates in prominent habitat types (Forget, Lambert, Hulme & Vander Wall, 2005). During field and laboratory trials in Hawaii, 10 of the 12 plant species investigated by Shiels and Drake (2011) had fleshy fruit

coverings and all but one native species had >44% of their seeds removed from the forest floor. Aside from *Prestoea*, the other three species in our study had 30–37% seed removal. While most of the species trialed in Shiels and Drake (2011) had their seeds destroyed when black rats interacted with them, the species with high seed survival were those with very small seeds (<1.5 mm) that passed intact through the black rat's digestive tract, and seed survival and dispersal of large-seeded species were rare. There is much evidence documenting how rodent seed removal often does not equate to seed predation (Forget et al., 2005; Shiels & Drake, 2011; Vander Wall, Kuhn & Beck, 2005), and our trials in the LEF were not set up to determine whether removed seeds were depredated or dispersed. Although seed dispersal by black rats may occur even when seeds are large (18–20 mm, like *Buchenavia*), the majority of the black rat interactions with seeds >1.5 mm result in seed predation because black rats are not known to store or scatterhoard seeds (reviewed in Shiels et al., 2014). Large-scale suppression or removal of invasive black rats from segments of the LEF for prolonged periods would be necessary to determine whether, and the extent to which, rats are altering the forest's plant community.

Our study confirms that black rats are present throughout all types of habitats (disturbed and undisturbed) within the LEF, and that forest gaps and stream gaps are sites where two of the four trialed tree species suffer greater seed removal by rats than in closed canopy forest. Even though seed removal by rats did not necessarily result in seed predation, frequent rat-seed interactions like we observed in the LEF and seed predation recorded elsewhere suggest that rats are likely altering recruitment of some plant species, thereby affecting community composition and forest structure. *Prestoea* seeds do not appear to be at much risk to rat predation in this forest. However, over one-third of the offered seeds of *Buchenavia*, *Guarea*, and *Tetragastris* were removed and most removals were due to black rats, which suggests that these plant species deserve further investigation to better determine whether black rats are affecting survival and recruitment of these populations. Furthermore, a species list that indicates the vulnerable species to predation by black rats would help inform management in the LEF and other tropical forests to better protect individual species or whole communities from the negative effects of black rats.

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## DATA AVAILABILITY

Data available from the Dryad Digital Repository: <https://doi.org/10.5061/dryad.tf806g8> (Shiels & Ramírez de Arellano, 2019) and the Luquillo Long-Term Ecological Research (LTER) Data Catalog (<https://luq.lter.network/datacatalog>).

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## REFERENCES

- Atkinson, I. A. E. (1985). The spread of commensal species of *Rattus* to oceanic islands and their effects on island avifaunas. In P. J. Moors (Ed.), *Conservation of Island birds* (pp. 35–81). Cambridge, U.K.: ICBP Technical Publication No. 3.
- Auld, T. D., Hutton, I., Ooi, M. K. J., & Denham, A. J. (2010). Disruption of recruitment in two endemic palms on Lord Howe Island by invasive rats. *Biological Invasions*, 12, 3351–3361.
- Banks, P. B., & Hughes, N. K. (2012). A review of the evidence for potential impacts of black rats (*Rattus rattus*) on wildlife and humans in Australia. *Wildlife Research*, 39, 78–88.
- Bennett, A. F. (1990). Habitat corridors and conservation of small mammals in a fragmented forest environment. *Landscape Ecology*, 4, 109–122.
- Brokaw, N., Crowl, T. A., Lugo, A. E., McDowell, W. H., Scatena, F. N., Waide, R. B. & Willig, M. R. (Eds.) (2012). *A caribbean forest tapestry*. Oxford, UK: Oxford University Press.
- Brokaw, N. V., & Grear, J. S. (1991). Forest structure before and after Hurricane Hugo in three elevations in the Luquillo Mountains, Puerto Rico. *Biotropica*, 23, 386–392.
- Capizzi, D., Bertolino, S., & Mortelliti, A. (2014). Rating the rat: Global patterns and research priorities in impacts and management of rodent pests. *Mammal Review*, 44, 148–162.
- Cox, M. P. G., Dickman, C. R., & Cox, W. G. (2000). Use of habitat by the black rat (*Rattus rattus*) at North Head, New South Wales: an observational and experimental study. *Austral Ecology*, 25, 375–385.
- Dowding, J. E., & Murphy, E. C. (1994). Ecology of ship rats (*Rattus rattus*) in a kauri (*Agathis australis*) forest of Northland, New Zealand. *New Zealand Journal of Ecology*, 18, 19–27.
- Drake, D. R., & Hunt, T. L. (2009). Invasive rodents on islands: Integrating historical and contemporary ecology. *Biological Invasions*, 11, 1483–1487.
- Forget, P.-M., Lambert, J. E., Hulme, P. E. & Vander Wall, S. B. (Eds.) (2005). *Seed fate: Predation, dispersal, and seedling establishment*. Wallingford, UK: CAB International.
- Graham, N. A. J., Wilson, S. K., Carr, P., Hoey, A. S., Jennings, S., & MacNeil, M. A. (2018). Seabirds enhance coral reef productivity and functioning in the absence of invasive rats. *Nature*, 559, 250–253.
- Harper, G. A., & Bunbury, N. (2015). Invasive rats on tropical islands: Their population biology and impacts on native species. *Global Ecology and Conservation*, 3, 607–627.
- Janzen, D. H. (1972). Association of a rainforest palm and seed-eating beetles in Puerto Rico. *Ecology*, 53, 258–261.
- Law, B. S., & Dickman, C. R. (1998). The use of habitat mosaics by terrestrial vertebrate fauna: Implications for conservation and management. *Biodiversity and Conservation*, 7, 323–333.
- Pimentel, D., Lach, L., Zuniga, R., & Morrison, D. (2000). Environmental and economic costs of nonindigenous species in the United States. *BioScience*, 50, 53–65.
- Reagan, D. P. & Waide, R. B. (1996). *The food web of a tropical rain forest*. Chicago, IL: University of Chicago Press.
- Richardson, B. A., Richardson, M. J., González, G., Shiels, A. B., & Srivastava, D. S. (2010). A canopy trimming experiment in Puerto Rico: The response of litter invertebrate communities to canopy loss and debris deposition in a tropical forest subject to hurricanes. *Ecosystems*, 11, 286–301.
- Shiels, A. B. (2010). *Ecology and impacts of introduced rodents (Rattus spp. and Mus musculus) in the Hawaiian Islands*. PhD Dissertation. Department of Botany, University of Hawaii at Manoa, Honolulu, HI.
- Shiels, A. B., & Ramírez de Arellano, G. E. (2019). *Data from: Habitat use and seed removal by invasive rats (Rattus rattus) in disturbed and undisturbed rainforest, Puerto Rico*. Dryad Digital Repository. <https://doi.org/10.5061/dryad.tf806g8>
- Shiels, A. B., & Drake, D. R. (2011). Are introduced rats (*Rattus rattus*) both seed predators and dispersers in Hawaii? *Biological Invasions*, 13, 883–894.
- Shiels, A. B., & Drake, D. R. (2015). Barriers to seed and seedling survival of once-common Hawaiian palms: The role of invasive rats and ungulates. *AoB Plants*, 7, plv057.
- Shiels, A. B., Flores, C. A., Khamsing, A., Krushelnycky, P. D., Mosher, S. M., & Drake, D. R. (2013). Dietary niche differentiation among three species of invasive rodents (*Rattus rattus*, *R. exulans*, *Mus musculus*). *Biological Invasions*, 15, 1037–1048.
- Shiels, A. B., & González, G. (2014). Understanding the key mechanisms of tropical forest responses to canopy loss and biomass deposition from experimental hurricane effects. *Forest Ecology and Management*, 332, 1–10.
- Shiels, A. B., Pitt, W. C., Sugihara, R. T., & Witmer, G. W. (2014). Biology and impacts of Pacific island invasive species. 11. *Rattus rattus*, the black rat (Rodentia: Muridae). *Pacific Science*, 68, 145–184.
- Shiels, A. B., & Ramírez de Arellano, G. E. (2018). Invasive rats (*Rattus* sp.), but not always mice (*Mus musculus*), are ubiquitous at all elevations and habitats within the Caribbean National Forest, Puerto Rico. *Caribbean Naturalist*, 48, 1–14.
- Shiels, A. B., Zimmerman, J. K., Garcia-Montiel, D. C., Jonckheere, I., Holm, J., Horton, D., & Brokaw, N. (2010). Plant responses to simulated hurricane impacts in a subtropical wet forest, Puerto Rico. *Journal of Ecology*, 98, 659–673.
- Snyder, N. F. R., Wiley, J. W., & Kepler, C. B. (1987). *The parrots of Luquillo: Natural history and conservation of the Puerto Rican parrot*. Los Angeles, CA: Western Foundation of Vertebrate Zoology.
- Towns, D. R., Atkinson, I. A. E., & Daugherty, C. H. (2006). Have the harmful effects of introduced rats on islands been exaggerated? *Biological Invasions*, 8, 863–891.
- Traveset, A., & Richardson, D. M. (2006). Biological invasions as disruptors of plant reproductive mutualism. *Trends in Ecology & Evolution*, 24, 208–216.
- Turvey, S. T., Oliver, J. R., Nargane Storde, Y. M., & Rye, P. (2007). Late Holocene extinction of Puerto Rican native land mammals. *Biology Letters*, 3, 193–196.
- Vander Wall, S. B., Kuhn, K. M., & Beck, M. J. (2005). Seed removal, seed predation, and secondary dispersal. *Ecology*, 86, 801–806.
- Weinbren, M. P., Weinbren, B. M., Jackson, W. B., & Villeda, J. B. (1970). Studies on the roof rat (*Rattus rattus*) in El Verde Forest. In H. T. Odum & R. F. Pigeon (Eds.), *A tropical rain forest: A study of irradiation and ecology at El Verde, Puerto Rico* (pp. E169–E182). Springfield, VA: U.S. Atomic Energy Commission, Division of Technical Information, National Technical Information Service.

- Willig, M. R., & Gannon, M. R. (1996). Mammals. In D. P. Reagan & R. B. Waide (Eds.), *The food web of a tropical rain forest* (pp. 400–429). Chicago, IL: University of Chicago Press.
- Wolf, C. A., Young, H. S., Zilliacus, K. M., Wegmann, A. S., McKown, M., Holmes, N. D., ... Croll, D. A. (2018). Invasive rat eradication strongly impacts plant recruitment on a tropical atoll. *PLoS ONE*, *13*, e0200743.

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