

Research

Pleistocene megafaunal extinctions and the functional loss of long-distance seed-dispersal services

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Pleistocene extinctions affected mainly large-bodied animals, determining the loss or changes in numerous ecological functions. Evidence points to a central role of many extinct megafauna herbivores as seed dispersers. An important step in understanding the legacy of extinct mutualistic interactions is to evaluate the roles and effectiveness of megafauna herbivores in seed dispersal. Here we use morphological and ecophysiological allometries to estimate both quantitative and qualitative aspects of seed-dispersal services likely provided by extinct megafauna. We developed a mechanistic model that encompasses four stages of seed dispersal – seed ingestion, gut retention, animal movement, and seed deposition. We estimate seed-dispersal kernels through simulations to infer the role of Pleistocene megafauna in promoting long-distance dispersal and examine how seed dispersal was affected by extinctions. Simulations suggest extinct large-bodied frugivores would frequently disperse large seeds over a thousand meters, whereas smaller-bodied frugivores are more likely to deposit the seeds over a few hundred meters. Moreover, events of long-distance seed dispersal by the extinct megafauna would be up to ten times longer than long-distance dispersal by smaller-sized extant mammals. By estimating the combined distribution of seed dispersal distances considering all large-bodied mammalian frugivores in specific South American Pleistocene assemblages we found that long-distance dispersal contracted by at least two thirds after the megafauna died out. The disruption of long-distance dispersal is expected to have consequences for recruitment, spatial and genetic structure of plant populations, population persistence and community composition. Promoting long-distance seed dispersal was one among other salient features of extinct Pleistocene megafauna that reveal their influence on natural ecosystems. Modeling the consequences of megafaunal extinctions can offer quantitative predictions on the consequences of ongoing defaunation to plant populations and ecological communities.

Introduction

The Late Quaternary extinctions (LQE) affected mostly large-bodied, terrestrial vertebrates, likely determining the loss of a sizable number of distinct ecological functions (Hansen and Galetti 2009, Bond 2010, Lopes dos Santos et al. 2013). Although most

studies have focused on the possible causes leading species to extinction (Koch and Barnosky 2006), more recent work has addressed the consequences of megafaunal extinction for ecological processes and ecosystem function (Doughty et al. 2013, Gill 2014, Smith et al. 2016). It is well known that large mammals are key elements of multiple ecosystem processes in modern ecological communities, including seed dispersal (Janzen and Martin 1982, Gautier-Hion et al. 1985, Bond et al. 2004, Burns 2014). The occurrence of large, overbuilt fruits in Central American dry forests led researchers to suggest that such fruit traits are the outcome of past selective pressures imposed by large, albeit extinct, megafauna (Janzen and Martin 1982). The so-called anachronic fruits have also been found in other megafauna-deprived ecosystems (Janzen 1986, Barlow 2000, Guimarães et al. 2008).

An important step in understanding the mutualism between megafauna and fruit-bearing plants and the consequences of losing these interactions is to examine the roles and the effectiveness of extinct megafauna as frugivores and seed dispersers. Besides the loss of several potential seed dispersers, which could by itself imply seed-dispersal limitation for some plant species, the megafaunal extinction truncated the body size distribution of mammals worldwide (Lyons et al. 2004, Dirzo et al. 2014), leaving just birds and a few smaller mammal species to carry on with seed-dispersal services (Hallwachs 1986, Guimarães et al. 2008, Pires et al. 2014). The seeds ingested by large mammals may be damaged during mouth or gut processing (Campos-Arceiz et al. 2012), may be deposited on unsuitable places for germination (O’Farrill et al. 2013), and are often dispersed in clumps, potentially resulting in greater competition and seed predation and decreasing the probability of recruitment as a consequence (Howe 1989, Fragoso 1997). Yet, seed dispersal by large mammals is key for certain plants, especially those that have seeds too large to be dispersed by smaller frugivores (Kitamura et al. 2002, Fragoso et al. 2003, Donatti et al. 2011, Sekar and Sukumar 2013). In fact, seed dispersal by large mammals may have provided an evolutionary route for fleshy-fruit plants to minimize the consequence of the trade-off between large seed size and seed-dispersal effectiveness (Guimarães et al. 2008).

Because large mammals can move over long distances (Carbone et al. 2005) they are more likely to promote long-distance dispersal and thus effectively contribute to connect plant populations across large spatial scales (Nathan et al. 2008). Moreover large mammals may have an important role in connecting seed-dispersal networks, thus coupling the dynamics of plant populations within a community (Vidal et al. 2013, Pires et al. 2014). Although all these general properties of seed dispersal by large animals are acknowledged, a necessary step to understand the legacy of megafaunal extinctions is to develop ways to quantitatively assess the consequences of the megafaunal losses for seed-dispersal services. Understanding the different effects of the LQE is paramount considering the on-going wave of vertebrate defaunation in the Anthropocene, which may have similar

or even greater consequences (Dirzo et al. 2014, Young et al. 2016).

Here we approach the problem of the functional consequences of megafaunal extinction to seed dispersal by addressing seed dispersal distance, a component of seed dispersal effectiveness (SDE; Schupp et al. 2010). We developed an agent-based model that estimates seed dispersal distance based on theoretical and empirical information on the scaling of seed ingestion, gut retention and animal movement with body mass. We then perform simulations to infer the contribution of extant and extinct large mammalian frugivores to seed dispersal. Our specific goal with such simulations was to obtain quantitative estimates on the frequency and scale of long-distance seed dispersal allowing us to compare seed-dispersal potential in past and modern mammal assemblages so as to better understand the ways whereby megafaunal extinctions might have affected plant populations and communities.

Material and methods

Allometric scaling and seed dispersal

Our rationale for modeling the functional role of extinct megafauna frugivores as seed dispersers is based on recent functional models of the movement ecology paradigm (Nathan et al. 2008), specifically in reference to the internal state (fruit consumption and handling, gut capacity and retention time) and movement. Both quantity and quality of seed dispersal can be affected by the body size of seed dispersers (Jordano 2000, Jordano and Schupp 2000, Pérez-Méndez et al. 2015). The amount of food consumed by an organism is determined by the time it spends foraging and the rate of food intake (Belovsky 1997). Considering organisms with similar dietary preferences, the time spent feeding on a certain food type should depend on the energy requirements of individuals, which are related to the three-quarter power of body mass ($\sim cM^{3/4}$), but is also limited by gut capacity, which increases in proportion to body mass (Van Soest 1996). The rate of food intake, in its turn, depends on the amount of food per bite, which is determined chiefly by mouth size, also increasing with body mass (Shiple et al. 1994, Van Soest 1996). Thus, even though the amount of food consumed relative to body mass declines with body size (Demment and Van Soest 1985, Owen-Smith 1988), the total amount of food consumed is greater for larger animals. Although different predictors of food intake have received support, suggesting the determinants of food intake are context dependent (Yearsley et al. 2001), a relationship between food intake and body mass is often acknowledged (Shiple et al. 1994). Thus, considering species with similar degrees of frugivory and assuming individuals of these different species spend about the same time feeding on fruits of a particular plant, individuals of larger species should consume a greater amount of fruits per unit time spent feeding on that particular plant.

Body size also affects the quality of seed deposition. Because energy requirements scale with the three-quarter power of body mass while gut capacity increases with body mass isometrically, larger animals are expected to have greater retention times (Van Soest 1996, Müller et al. 2013). Moreover larger animals have larger home ranges (Harestad and Bunnell 1979, Jetz 2004) and are expected to travel farther daily (Carbone et al. 2005). Thus, seeds ingested by large-bodied mammals should have a greater probability of being deposited far from the maternal plant when compared with seeds ingested by small-bodied mammals, which should, on average, increase the likelihood of recruitment (Terborgh et al. 2008, Schupp and Jordano 2011).

To evaluate the potential of extinct Pleistocene herbivores as seed dispersers we first compiled a list of extinct North and South American Pleistocene mammals likely to include fruits in their diet and their estimated average body mass (Supplementary material Appendix 1 Table A1). We consider only Pleistocene mammals weighting more than 10 kg, which would be more likely to disperse seeds of large-seeded plants. This definition allows us to consider both large mammals that survived the Quaternary extinctions and the extinct megafauna, usually defined as mammals with body mass > 44 kg (~100 lb; Koch and Barnosky 2006). We then compiled data (Supplementary material Appendix 1 Table A2) on the relationship between body mass of extant mammals and the components affecting seed-dispersal distance: gut capacity as a measure of food intake (Belovsky 1997), mean retention time (Owen-Smith 1988, Van Soest 1996), and daily range as a measure of movement ability (Carbone et al. 2005). We performed linear regression tests using the compiled data for extant mammals so we could infer the parameters and prediction intervals of linear models and estimate the expected values and range of gut capacity, mean retention time and daily range for the extinct megafauna.

We applied here the 'biological uniformitarianism' rule and use morphological features of known ecological function in the present day as indicators of functional relations in the past (Tiffney 2004). Biological uniformitarianism is a fundamental assumption of all paleoecological studies and we assume the scaling rules described above, which have both empirical and theoretical support, also applied for mammals that died out a few thousands of years ago. Along this line of reasoning, we used the prediction interval for the regressions instead of the confidence interval for the slopes in order to get more conservative estimates of inference errors for the specific allometric trends examined and the inferences obtained from them.

Modeling seed dispersal

To test how frugivores of different body sizes would compare in terms of seed-dispersal distance we developed an agent-based model that simulates seed dispersal of a large-seeded plant species in a two-dimensional landscape (Fig. 1). In this approach, we simulate the process of tracking seeds ingested from a plant or plant aggregation in one feeding

event until their elimination. Then, we estimate the resulting seed-dispersal kernels (Westcott et al. 2005, Dennis and Westcott 2007). Seed-dispersal kernels are probability functions depicting the location of seed deposition relative to the source (Nathan 2006, Nathan et al. 2012). Building seed-dispersal kernels requires estimates 1) of the number of removed seeds, 2) of how long seeds are retained, and 3) how far from the source dispersers move during that time (Dennis and Westcott 2007).

In our model, the number of seeds ingested by individuals of a given species in one foraging event is determined by the estimated species-specific gut capacity, which in turn depends on body size. For simplicity we assumed species have the same degree of frugivory so that the proportion of the gut capacity occupied by seeds (20% of the diet in the baseline simulations) was similar. There is evidence that fruits may constitute a smaller fraction of the diet of very large herbivores than in small or medium-sized ones, albeit the nature of this relationship is not well understood. Therefore, we also performed a sensitivity analysis where we relaxed the assumption used in the baseline simulations and randomly sampled the proportion of fruits in the diet of each species from a uniform distribution ranging between 0.1 and 0.7, thus exploring a number of combinations of frugivory degree for species of different body sizes (Supplementary material Appendix 1 Fig. A1). The fraction of gut capacity comprised by fruits was divided by the average mass of fresh fruits of type I megafauna plants (~150 g; Guimarães et al. 2008) to determine the average number of seeds ingested per species. Type I megafauna plants are those bearing large fleshy fruits (diameter: 4–10 cm) with a few (≤ 5) large seeds (Guimarães et al. 2008). The number of seeds ingested per individual was then sampled from a Poisson distribution with parameter λ equal to the average number of seeds ingested per species. Note we are focusing here on endozoochory, although seeds can also be dispersed by other means such as spitting (Bodmer 1991), epizoochory (Couvreur et al. 2008) and transport by secondary seed dispersers (Donatti et al. 2009).

The deposition times of ingested seeds are drawn from a Gamma distribution representing seed retention in the gut (Guttal et al. 2011). Each sampled value corresponds to the retention time of one ingested seed. We used the Gamma distribution because of its versatility, which allows reproducing different forms of one-tailed distributions. Moreover the Gamma distribution can be easily parameterized since its parameters, shape (k) and scale (θ), can be defined in terms of the empirical mean (\bar{t}) and variance (s^2) of seed retention time (Guttal et al. 2011):

$$k = \frac{\bar{t}^2}{s^2} \quad (1)$$

$$\theta = \frac{s^2}{\bar{t}} \quad (2)$$

We use the relationship between mean seed retention time and species body mass (Fig. 1) to define \bar{t} and set the variance to $s^2 = 400$. By setting a large variance relative to the

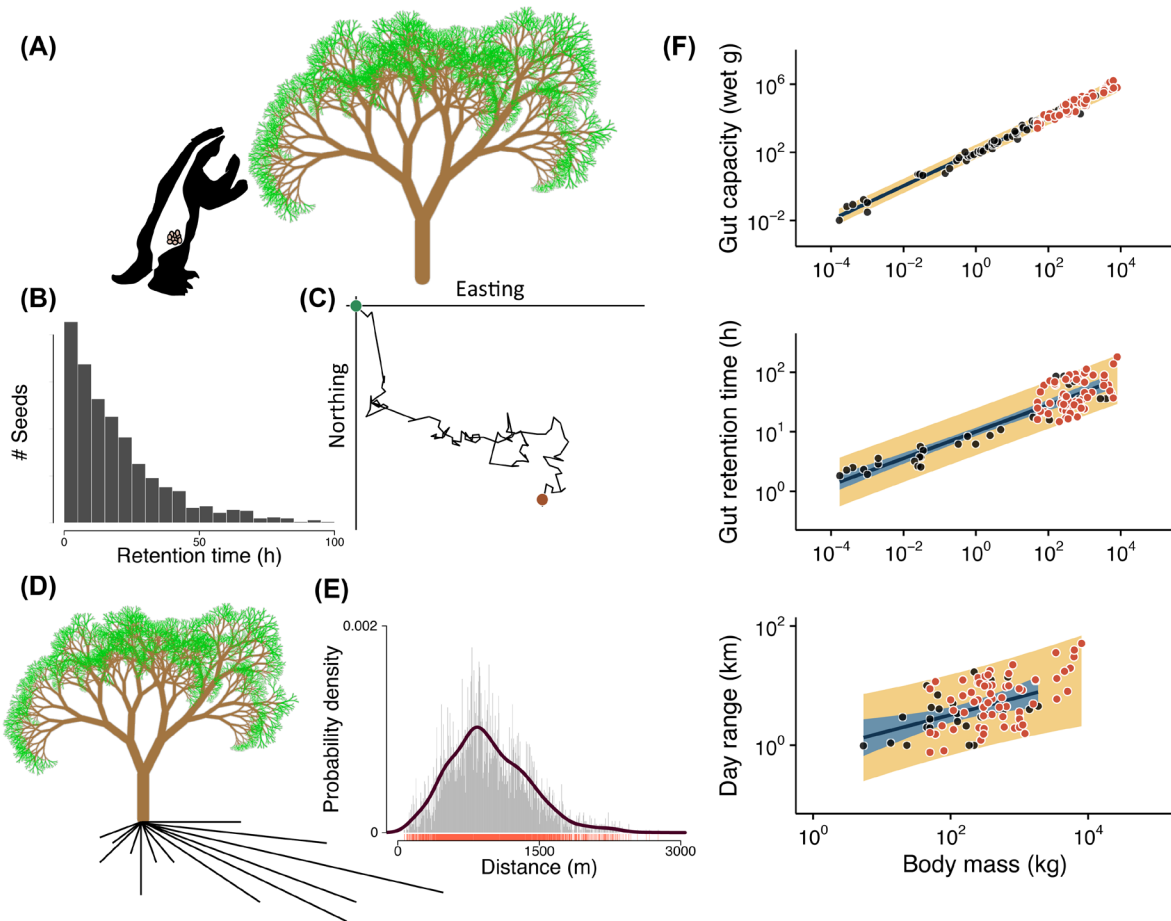


Figure 1. Modeling seed dispersal as a multistage process. (A) Seed ingestion is constrained by the gut capacity of the frugivore, here illustrated by a giant sloth (*Megatheriidae*). (B) Seed retention is modeled as a fat-tailed distribution representing the relative frequency of seeds eliminated at different times after ingestion. (C) Animal movement is simulated under models where individuals can move in any direction after ingesting the seeds of a plant or plant aggregate. The trajectory from the starting (green dot) to the final position (brown dot) is depicted as a solid line. (D) While moving, the frugivore deposits the seeds at different distances from the source. (E) Seed dispersal kernels – probability density functions of dispersal distances – are estimated by combining information on retention time of ingested seeds and movement. (F) Empirical relationships between variables related to the processes illustrated in panels (A–C) and body mass. Black dots represent compiled data for extant mammals. Shaded regions depict the confidence (blue) and prediction (yellow) intervals for the regression. Colored points represent extinct megafauna species. For extinct species, the positions on the y-axis are random locations within the prediction interval estimated from the species-specific estimated body mass (x-axis).

mean, the seed retention time distribution will have fat tails, but the maximum retention times are within a realistic range according to previous seed-dispersal studies with extant large frugivores, i.e. maximum retention time ~ 250 h, used here as an upper bound for the very large herbivores (Dinerstein and Wemmer 1988, Campos-Arceiz et al. 2008, 2012). Because the relationship between mean retention time and body mass has been challenged (Steuer et al. 2011) we also ran simulations assuming all species have similar mean retention times (30 h). The results are qualitative similar to those obtained with the baseline simulations (Supplementary material Appendix 1 Fig. A2).

We simulated the process of tracking movements of individuals for a fixed amount of time by measuring the distance an individual moved after consuming fruits and ingesting the

seeds of plants at a certain point in two-dimensional space (Westcott and Graham 2000). We simulated animal movement under four different models: 1) the Brownian walk (BW) where individuals move randomly in both distance and direction (Bartumeus et al. 2005); 2) the correlated random walk (CRW) where individuals move erratically towards a specific direction (Auger-Méthé et al. 2015), which could represent another food source; 3) the Lévy walk model (LW), where most of the movements are over random short distances, but longer ‘steps’ eventually take place (Bartumeus et al. 2005, Benhamou 2007); 4) the composite correlated random walk (CCRW), which consists in a combination of two movement behaviors: an intensive search phase with more restricted movements and an extensive phase corresponding to nearly straight long distance steps (Auger-Méthé

et al. 2016). Therefore the BW is the most conservative model in terms of traveled distance, whereas the others tend to generate movement patterns with greater spread. We present here the results on this most conservative scenario, but the results for other models are presented as Supplementary material Appendix 1. Note that although the CCRW model has been shown to be more accurate in describing the movement of mammals in the scale of years (Auger-Méthé et al. 2016), here we simulate movement over a few days and the other models may be reasonable approximations at this scale. For instance, recent critics of Lévy walk models (Pyke 2015) argued for its potential value when implementing individual-based models of movement in terms of both theory and observations, especially in relation to distribution, dispersal and other population-level phenomena.

Because travel distance is related to body size, we assume body mass affects the expected step length in the models of animal movement. In the BW, CRW and CCRW step length can be defined by the minimum step length (a) plus an increment sampled from an exponential distribution with rate parameter λ (Auger-Méthé et al. 2015). Because the mean of the exponential distribution approximates λ^{-1} , we define $a = 0$ and $\lambda = cd^{-1}$, where c is some constant and d is the expected travel distance per hour, calculated from the relationship between daily range and body mass (see Supplementary material Appendix 1 Table A3 for further details on model parameters). Lévy walks were simulated with $a = cd^{-1}$ and exponent $\mu = 2.95$ to keep the body mass scaling relationship while avoiding exceedingly long steps (Supplementary material Appendix 1 Table A3). We performed all simulations and analysis in R (R Core Team). We used the *adehabitat* R package (Calenge 2006) and functions from Auger-Méthé et al. (2015) to simulate individual movement. We assume time steps are measured in hours instead of seconds as in the functions of the *adehabitat* R package. The functions used to perform simulations and plot seed-dispersal kernels are available as Supplementary material Appendix 1 Data A1.

With information on how far from the source individuals would be at each hour and the deposition times of ingested seeds we estimated seed-dispersal distance for each ingested seed (Fig. 1). Following Pérez-Méndez et al. (2016) we define long-distance dispersal (LDD) events as those beyond the 95th percentile of the distribution of seed-dispersal distances. We thus examine how seed-dispersal kernels and the scale of LDD change for a large-seeded plant dispersed by mammalian frugivores of different body-sizes. To test the contribution of each variable to seed-dispersal kernels we also ran an ancillary set of analyses in which only one variable scaled with body mass at a time, while the remaining variables were assigned the same value for all species.

The contribution of each frugivore species to the seed-dispersal events in a given locality also depends on its population size. Larger-bodied species often occur in lower densities when compared to smaller animals. The estimated decline of population density with increasing body

mass also suggests a $3/4$ exponent (Damuth 1981), presumably related to the scaling of resource use and resource limitation (Brown et al. 2004). Although this scaling relationship – termed Damuth’s law – can predict global scale patterns, body size may explain only a small part of the variation in local abundances (White et al. 2007). For this reason it has been suggested that Damuth’s law may be used to infer an upper boundary on local abundances which are context dependent (White et al. 2007). To ensure all species are represented by at least one individual we scale the area considered in simulations accordingly (10 km^2).

Studied assemblages

Using the modeling approach described above we projected seed-dispersal kernels and estimated the threshold defining LDD events for species within different ranges of body sizes. We performed 100 simulations for each species to encompass uncertainty. To investigate how megafaunal extinction changed the potential for seed dispersal of large-seeded plants we simulated seed dispersal by populations of each of the large mammalian species present in five different regions in South America during the late Pleistocene: southern Brazil (França et al. 2015), São Raimundo Nonato in northeastern Brazil (Guérin 1991), central Chile (Encina 2015), Santa Elena peninsula in Ecuador (Lindsey and Lopez 2015) and the Pampean region in Argentina (Fariña et al. 2014). Data for each assemblage is summarized in Supplementary material Appendix 1 Table A4. We consider for these simulations only those species that were identified as browsers or mixed-feeders in paleoecological studies (MacFadden et al. 1994, França et al. 2015), being thus more likely to include fruits in their diets and act as seed dispersers. We also estimated seed-dispersal kernels, under the same set of assumptions, considering the large mammalian frugivores present in the Brazilian Pantanal (Donatti et al. 2011). The Brazilian Pantanal bears a highly diverse mammalian assemblage when compared to other biomes in South America, including the largest extant frugivore, the lowland tapir *Tapirus terrestris*, which disperses the seeds of many large-seeded plants (Galetti et al. 2001, Fragoso et al. 2003, O’Farrill et al. 2013). For most of the analyzed regions, we have limited evidence on the early Holocene frugivore assemblages resulting from the late Pleistocene extinctions. We thus use the Pantanal mammalian assemblage for comparison here as a best-case scenario, in terms of post-extinction mammalian diversity, in order to obtain conservative estimates of seed-dispersal limitation after extinctions.

To quantitatively assess the potential for seed dispersal in each of these localities we combined the seed-dispersal kernels resulting from simulations for the different species to build total seed-dispersal kernels (Nathan et al. 2012). One important caveat about these simulations is that several ungulates such as peccaries (Tayassuidae) and cervids (Cervidae), besides acting as seed dispersers (Gautier-Hion et al. 1985, Bodmer 1990, Prasad et al. 2006), are also known

to damage seeds, behaving as seed predators for many plant species (Bodmer 1991). However, we do not know to which extent this also applies for extinct large mammals. Therefore we assume for simplicity that the amount of damaged seeds is comparable in all species considered. Because differences in seed processing would change the effective number of dispersed seeds, we address this limitation through the sensitivity analysis where the degree of frugivory was assigned at random across species. However, we stress that our goal with our simulations is not to obtain highly accurate estimates for the deposition distance of intact seeds and seed-dispersal kernels in the past, but to infer the potential for seed dispersal in a comparative framework and how it would have changed after the megafaunal extinctions.

Results

The body mass distribution of non-arboreal frugivores in the Americas is considerably different when examining only extinct (median = 390 kg; range: 50–8000 kg; Supplementary material Appendix 1 Table A1) or only extant species (median = 55.5 kg; range: 16–360 kg; Supplementary material Appendix 1 Table A1). Because body mass is linearly related (in the log-log scale) to most variables affecting seed-dispersal distance – gut capacity ($a = 2.031$, $b = 1.002$, $r^2 = 0.98$, $p < 0.001$), retention time ($a = 1.001$, $b = 0.224$, $r^2 = 0.88$, $p < 0.001$) and movement ability ($a = -0.092$, $b = 0.300$, $r^2 = 0.28$, $p < 0.001$) – extinct Pleistocene mammals were at the highest extreme of the distribution of each of those variables (Fig. 1F). For instance, considering species with similar degrees of frugivory, frugivores with mean body mass $\sim 10^3$ kg should remove about 10 times more seed biomass than frugivores weighting $\sim 10^2$ kg. The large frugivores would be expected to retain seeds in the gut for periods on average 70% longer than the smaller species. Finally, a 10^3 kg mammal is expected to travel daily distances twice as large as distances traveled by a 10^2 kg species (Fig. 1F).

Our simulations of seed dispersal show that differences in the body mass of seed dispersers should result in disparate seed-dispersal kernels (Fig. 2). Events of long-distance dispersal should be between 4 and 10 times longer for a 10^3 kg mammal when compared to a mammal between 10 and 50 kg (Fig. 2), depending on the movement model (Supplementary material Appendix 1 Fig. A3–A5). This means that while any dispersal event farther than a few hundred meters could be considered a LDD event for a 50 kg mammal, seed dispersal by a megaherbivore, such as a gomphothere, would very frequently exceed hundreds of meters and LDD events would be those above a few thousand meters (Fig. 2). Ancillary analyses where only one of the variables – gut capacity, mean retention time, or travel distance – was assumed to scale with body mass show that variation in gut capacity has minimal impact on the shape of seed-dispersal kernel. The effect of variation on gut capacity would be mainly quantitative, impacting the absolute frequency of seeds deposited at different distances. Conversely, variation in retention time

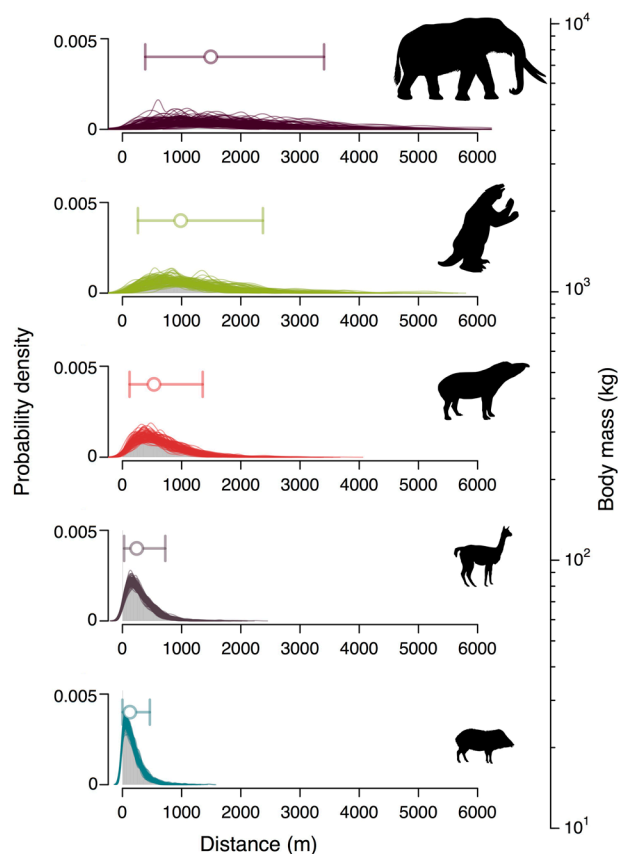


Figure 2. Seed-dispersal kernels depicting seed dispersal of a large-seeded plant by populations of mammalian frugivores within different body size ranges. Animal movement was simulated as a Brownian walk (BW). Each line represents one of 100 simulations. Each kernel panel is located within the approximate body mass range used in the simulation. Circles represent the median and bars determine the 5th and 95th percentiles. The 95th percentile value defines the minimum threshold value considered a LDD event.

and especially in travel distance result in disparate kernels for frugivores with different body mass (Supplementary material Appendix 1 Fig. A6).

Our simulations of the combined species-specific seed-dispersal kernels for different mammalian assemblages show that a large-seeded plant in the past should have had consistently high probabilities of dispersal over distances in the order of kilometers (Fig. 3). In contrast, the total seed-dispersal kernel in the modern assemblage used here as a benchmark, the Pantanal, is highly skewed towards short distances and seed-dispersal distance rarely exceeds 1000 m (Fig. 3, Supplementary material Appendix 1 Fig. A7–A9). The frequency of dispersal events within a particular range can be estimated from the area under the kernel. While our simulations using the most conservative movement model (BW) suggest that only about 5% of the seeds ingested by large mammals in the Pantanal would be dispersed for distances > 1000 m, the percentage of seeds being dispersed over the same distance and above in the analyzed Pleistocene assemblages would be higher than 40%.

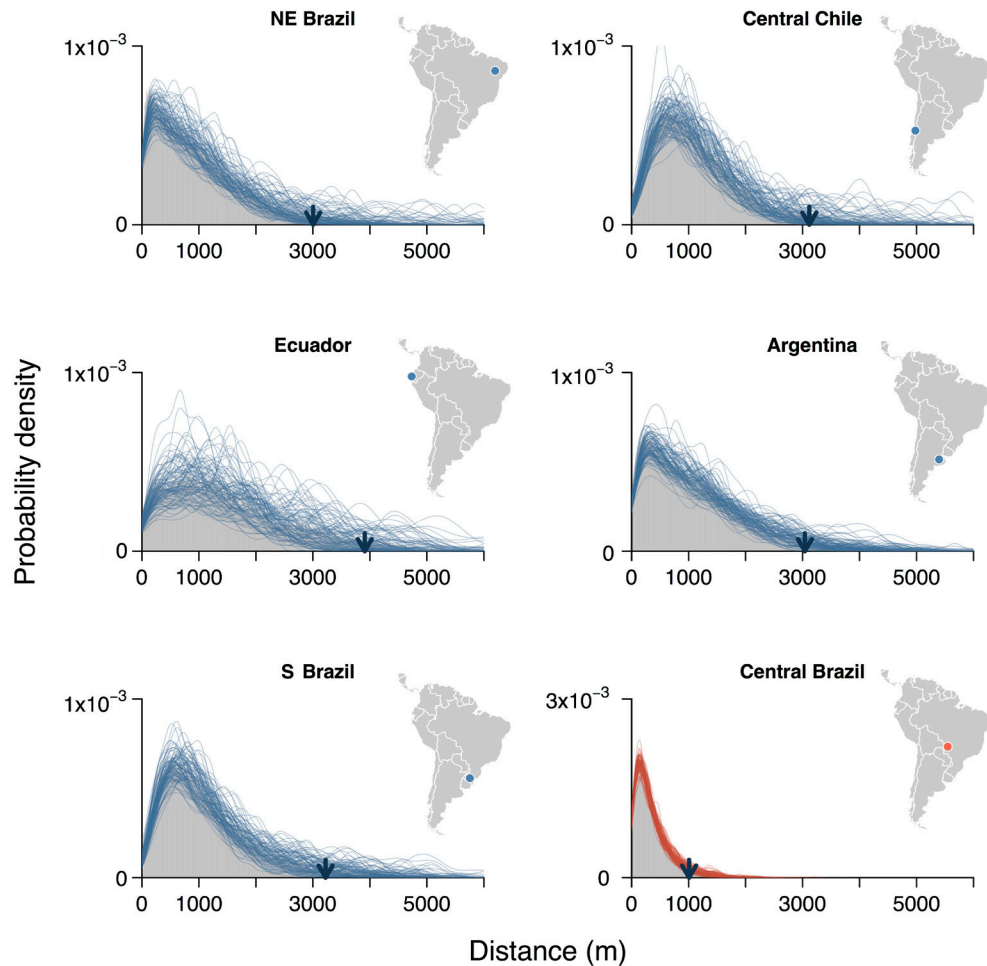


Figure 3. Total seed-dispersal kernels depicting seed dispersal of a large-seeded plant by large mammals in past (blue) and modern (red) mammalian assemblages representing specific locations. Animal movement simulated as a Brownian walk (BW). Each line represents one of 100 simulations. Arrows show the 95th percentile, which defines the magnitude of LDD events. Approximate locations of the six analyzed sites are depicted in the inset maps.

Discussion

The estimates of variables related to dispersal distance for the Pleistocene megafauna suggest that mammals such as ground sloths and gomphotheres would have been unique in terms of the amount of seeds mobilized and how far they could move the seeds. Accordingly, our simulations indicate that long-distance seed dispersal was considerably hindered by megafaunal extinctions and that extant frugivores are unlikely to compensate for the loss of the unique megafaunal seed-dispersal services.

We simulated seed dispersal under multiple assumptions regarding functional aspects of fruit consumption, retention times, population densities, and movement patterns. Unfortunately, there is no alternative when dealing with extinct species for which functional aspects such as those considered in our analysis can only be inferred. Given that the allometric scaling is consistent for the large megaherbivores considered (our main underlying assumption), the inferences about the trends should be reasonably robust. We also chose not to

include in the model additional parameters that could add further complexities such as social behavior, habitat preferences and habitat heterogeneity, all of which can affect seed deposition patterns (Fragoso 1997, Karubian and Durães 2009). The simplicity of the proposed model, and the fact that each of the stages is modeled independently, allows one to explore variations of such assumptions in future work. Despite such simplifications, seed-dispersal distances obtained from simulations are within the range found for extant megaherbivores such as elephants (Blake et al. 2009, Campos-Arceiz and Blake 2011, Bunney et al. 2017), suggesting that the assumed relationships between body size and model variables allow realistic inferences of seed-dispersal kernels. For instance, using the relationship between body mass and seed-dispersal related variables we estimated that the distance of seed dispersal by gomphotheres, the largest South American Pleistocene mammals, would very frequently exceed a few kilometers, with LDD events greater than 3.5 km. Campos-Arceiz et al. (2008) found Asian elephants *Elephas maximus* in Sri Lanka disperse seeds on average between 1 and 2 km

and recorded events of LDD > 6 km. African elephants *Loxodonta africana* in Congo (Blake et al. 2009) and in South Africa (Bunney et al. 2017) were found to disperse most of the ingested seeds to distances above 1 km, with maximum recorded distances > 60 km. Our simulations indicate similar trends for many extinct megafauna, with high frequency of dispersal events in the scale of kilometers and potential for even larger extreme values for LDD events.

By simulating seed dispersal by extinct mammals in specific Pleistocene assemblages and comparing with a modern species-rich assemblage we show that the loss of large frugivores most certainly impaired LDD for large-seeded plants. Importantly, by using a conservative model we obtain a quantitative estimate of the minimum impact of megafaunal extinction on seed-dispersal effectiveness, suggesting that the magnitude of LDD would have shrunk substantially. Even in habitats with a species-rich mammal assemblage such as in the Pantanal (Donatti et al. 2011), large-seeded plants are expected to have considerably shorter seed-dispersal distances when compared to the Pleistocene mammalian assemblages. This highlights a baseline shift after megafaunal extinction (Corlett 2013): what we currently consider LDD would be considered short-distance dispersal in the past. Therefore, we argue that the loss of megafaunal seed dispersers led to a scale shift in the seed-dispersal kernels, reducing LDD by at least two thirds, in our most conservative scenario.

Insufficient LDD may have a suite of ecological and evolutionary consequences that ultimately shape the distribution of plant species and the composition of ecological communities (Trakhtenbrot et al. 2005). Long-distance dispersal limitation, for instance, may impact population persistence by increasing the effects of negative density dependence across the different plant life stages (Fragoso 1997, Caughlin et al. 2014, Pérez-Méndez et al. 2015), disrupting the rescue effects associated with connectivity among forest patches and metapopulation dynamics (Cain et al. 2000). The limitation of dispersal distance also reduces the chance of rapid shifts in plant distribution (Trakhtenbrot et al. 2005), which may have limited the distribution of some plant species (Doughty et al. 2016) to regions that became climatically suboptimal as climate changed during late Pleistocene and the Holocene. Future studies may help us understand the consequences of Pleistocene mammalian extinctions to plant populations and plant communities by investigating how the distribution of large-seeded plants changed over time.

One important caveat is that we focus here on endozoochory. However, seed-dispersal services promoted by secondary seed dispersers such as scatter-hoarding rodents may be important for some plants species (Jansen et al. 2012). In regions where scatter-hoarding rodents are overabundant, secondary seed dispersal and intensive pilfering may result in considerably long seed-dispersal events (Jansen et al. 2012), but still well below the frequency and distances attained by large mammals as suggested by empirical data and by our estimates for extinct Pleistocene megafauna. In addition,

habitat loss and fragmentation also affect some of these smaller-sized species, impacting their role in dispersing the seeds of large-seeded plants (Galetti et al. 2006, Donatti et al. 2009).

The reduction of LDD after megafaunal extinctions likely changed the genetic structuring of plant populations over time (Collevatti et al. 2003) by reducing the connectivity of large-seeded plant populations in heterogeneous landscapes (Carvalho et al. 2015). If so, we should expect that plants primarily dispersed by large mammals show greater genetic structuring among populations when compared to plant species with fully functional dispersal modes. Future studies should focus on examining relative amounts of genetic differentiation among populations to test whether fruiting plants with the megafaunal dispersal syndrome (Guimarães et al. 2008) have greater genetic differentiation than those dispersed by smaller animals or abiotic means. Gene flow between plant populations depends on both seed dispersal and pollination. Although pollination is thought as the chief process promoting gene flow, the importance of seed dispersal is being increasingly appreciated and may be more important than pollen-mediated dispersal for many species (Dick et al. 2008, García and Grivet 2011, Carvalho et al. 2016), especially in connecting otherwise genetically isolated populations (Jordano et al. 2007). On the other hand, pollination may partially compensate the loss of large seed dispersers promoting gene flow among subpopulations (Pérez-Méndez et al. 2016). Studies separating these confounding effects by examining different markers may shed light on the consequences of Pleistocene extinctions to the genetic structure of plant populations.

Our findings using a spatial model to quantify the role of megafauna as seed dispersers are especially relevant when considering the ongoing defaunation, which extirpates species across the globe. The most vulnerable animals are often the largest (Galetti and Dirzo 2013, Dirzo et al. 2014) and the effects of losing large frugivores can already be seen in some places where plants dispersed by smaller dispersal agents or abiotic means are becoming overrepresented (Wang et al. 2007, Wright et al. 2007, Harrison et al. 2013). Seed-dispersal services may be partially compensated by other potential seed dispersers, including birds and smaller mammals (Jansen et al. 2012), or by haphazard, sporadic dispersal by gravity or runoff (Pérez-Méndez et al. 2016). However, this does not seem to be the case for many large-seeded plants, as several studies suggest that the surrogate seed disperser may not be as effective as the largest ones (Poulsen et al. 2002, Donatti et al. 2009, Campos-Arceiz et al. 2012, Bueno et al. 2013, Kistler et al. 2015, Sekar et al. 2015). Modeling approaches like the one presented here and in other studies on seed dispersal (Doughty et al. 2016) can be a useful tool to examine how biodiversity loss impacts ecosystem processes and, more specifically, to what extent surrogate species or other seed-dispersal mechanisms compensate the loss of seed-dispersal services. While many knowledge gaps still remain to uncover some aspects of

seed-dispersal effectiveness by extinct animals, our exercise highlights salient features of Pleistocene megafauna that help revealing their enormous influence in their habitats and understanding current traits and patterns shown by their plant partners surviving in present-day environments.

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Supplementary material (Appendix ECOG-03163 at <www.ecography.org/appendix/ecog-03163>). Appendix 1.