Article

Latitudinal gradients in seed predation persist in urbanized environments

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Urbanization is creating a new global biome, in which cities and suburbs around the world often resemble each other more than the local natural areas they replaced. But while urbanization can profoundly affect ecology at local scales, we know little about whether it disrupts large-scale ecological patterns. Here we test whether urbanization disrupts a macroecological pattern central to ecological and evolutionary theory: the increase in seed predation intensity from high to low latitudes. Across 14,000 km of latitude spanning the Americas, we compared predation intensity on two species of standardized experimental seeds in urbanized and natural areas. In natural areas, predation on both seed species increased fivefold from high latitudes to the tropics, one of the strongest latitudinal gradients in species interactions documented so far. Surprisingly, latitudinal gradients in predation were equally strong in urbanized areas despite significant habitat modification. Nevertheless, urbanization did affect seed predation. Compared with natural areas, urbanization reduced overall predation and vertebrate predation, did not affect predation by invertebrates in general, and increased predation by ants. Our results show that macroecological patterns in predation intensity can persist in urbanized environments, even as urbanization alters the relative importance of predators and potentially the evolutionary trajectory of urban populations.

Biologists have long speculated that interactions among species become stronger towards the tropics[1,2]. This latitudinal gradient in increasing interaction intensity, paralleling gradients in temperature, primary productivity and biodiversity, is thought to have played a major role in shaping global patterns in ecology and evolution[3–5]. For example, increasing risk of being eaten from high to low latitudes is invoked to explain long-distance migration[6], species' equatorward range edges[7], the astounding diversity of tropical forests[8,9] and faster speciation in the tropics[10]. Large-scale experiments have now demonstrated many dramatic latitudinal gradients in the intensity of predation, both within and across terrestrial and coastal biomes and at multiple trophic levels[6,11–13] (reviewed in ref. 2). However, it remains unclear how robust such macroecological patterns are to accelerating anthropogenic change.

One of the most intense and rapidly expanding forms of land use change is urbanization[14,15]. Urbanization's ecological effects can be seen even at its earliest stages, as natural vegetation in rural areas is cleared to make way for human homes[16]. As urbanization intensifies, many

species decline in abundance or disappear altogether[17,18], although a few experience population booms[19,20]. This biotic reshuffling can alter the nature and intensity of local trophic interactions[21–23], impacting ecosystem services, ecological functioning, and evolution in cities and their suburbs[24–26]. But urbanization is more than a patchwork of local effects; it has created a novel biome in which urbanized areas often resemble other, distant urbanized areas more than adjacent natural areas[27,28]. While local effects of urbanization are well studied, we know little about whether macroecological patterns are maintained in this new global biome[29,30].

Here we test whether the homogenizing effect of urbanization flattens latitudinal gradients in predation intensity. We consider post-dispersal seed predation, a globally important interaction that doubles in intensity from the Arctic to the equator, at least in the Americas[13,31]. Seed predation kills the first independent life stage of plants (as opposed to herbivory, which plants generally survive), and thus strongly affects plant demography, community assembly, geographic

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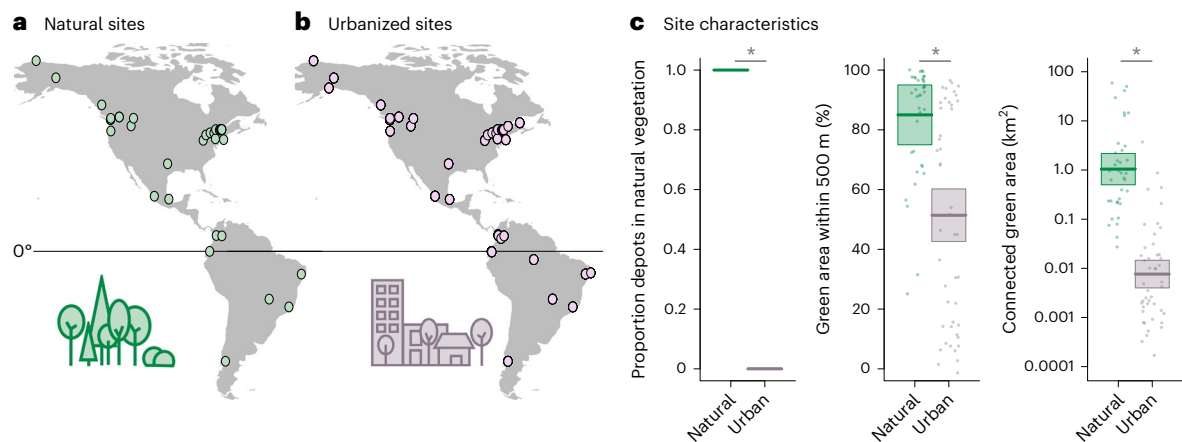
Fig. 1 | Site locations and characteristics. a,b, Locations of experimental sites in natural areas (**a**; *n* = 36 sites) and in urbanized areas (**b**; *n* = 45). **c**, Compared with natural sites, urbanized sites had less ground surface with untended, natural vegetation and litter (left); less green area within a 500 m radius (middle; green areas defined as those not covered with buildings, paving or stonework); and smaller connected green areas (right; note the log scale on *y* axis). Horizontal lines, boxes and points show the mean, 95% CI and partial residuals extracted from models with fixed effects 'urbanization + absolute latitude', 1 measurement per site. * denotes significant differences between natural and urbanized areas. None of the three response variables varied with latitude (statistical results in Extended Data Table 1), so all are plotted for the median absolute latitude (45°). CI and residuals in left plot are too narrowly distributed around the mean to be visible.

distributions and evolution[9,32–34]. While most theory proposing latitudinal gradients in predation does not specify a trophic level[4,10], stronger seed predation towards the tropics is proposed to both ecologically maintain and evolutionarily accelerate the spectacular diversity of tropical plants and their enemies[8,9,35]. Further, urbanization can strongly affect seed fates, leading to rapid evolution[36]. As the urban biome is set to double in size by 2100[37], its effects on the macroecological patterns that shape biodiversity, such as latitudinal gradients in predation, are increasingly important to the future distribution and evolution of life.

One might expect urbanization to flatten latitudinal gradients by reducing latitudinal differences among habitats. Grassy lawns in Brazil and Canada resemble each other far more than the tropical and boreal forests they replaced ('biotic homogenization'; see Methods). Indeed, both abiotic conditions[16] and species assemblages[38,39] are more similar among urban areas than rural ones. Urbanization can flatten latitudinal gradients in species diversity[30,40,41], which are proposed to both drive and reflect latitudinal gradients in species interactions[3]. However, the few empirical studies to test among potential mechanisms for latitudinal predation gradients find that predation rates are only loosely related to predator diversity[13,31,42,43]. Indeed, predation rates should more directly reflect predator abundance or per capita foraging intensity, which do not necessarily change in synchrony with diversity[2]. If the overall abundance or foraging intensity of predators is maintained in urbanized environments[24,44,45], urbanization may not affect predation rates even if it reduces biodiversity. Or, if urbanization reduces predator abundance similarly across latitudes, urbanized sites may have lower predation but still maintain latitudinal gradients in predation intensity.

Using consistent experimental methods and >56,000 seeds, we compared latitudinal gradients in predation between urbanized sites (urban and suburban backyards) versus nearby natural areas across 112° of latitude (Fig. 1). This work is an extension of the Biotic Interaction Gradients (BIG) experiments, which found steep latitudinal clines in seed predation when measured in large wilderness areas[13,31]; here we test whether these gradients are maintained in smaller natural areas and in urbanized areas.

Two key challenges in comparing predation rates across large spatial gradients are the lack of shared prey species among disparate communities and the potential for anti-predator adaptations in high-predation environments to mask the underlying latitudinal patterns of interest[46,47]. To meet these challenges, we measured predation on agricultural seeds that have been bred for human consumption and therefore possess minimal chemical or physical defenses. As the effects of urbanization vary dramatically among animal taxa[48], we distinguished among three types of predation: total predation, measured using sunflower seeds without shells, which are eaten by diverse invertebrates and vertebrates; invertebrate predation, measured by caging some sunflower seeds to exclude vertebrate predators; and vertebrate predation using oats with their thin hulls intact, which are eaten almost exclusively by small mammals and birds (see Methods).

We measured seed predation at 81 sites from the Arctic tundra to southern South America (Fig. 1). We set seeds out in small piles ('depots') on the ground in areas with vegetated ground cover, be it lawns, garden beds, or untended areas with natural litter. After 24 h, we quantified predation and noted any indicators of predator identity (for example, invertebrates still eating seeds, rodent faeces). We ran the experiment 1 to 11 times per site, pairing experiments at urbanized and nearby natural sites within a few days of each other when possible.

Results and Discussion

We found strong latitudinal gradients in seed predation but little evidence that these gradients are flattened by urbanization. Across 337 runs of the experiment at 81 study sites, seed predation increased more than fivefold from polar latitudes to the equator (Fig. 2). Latitudinal gradients in predation did not differ between urbanized and natural areas (latitude × urbanization: $x^2_{\text{d.f.}=1} = 0.11$, $P = 0.74$; Fig. 2), and this was true for total seed predation, predation by invertebrates only and predation by vertebrates (latitude × urbanization × predation type: $x^2_{\text{d.f.}=2} = 1.15$, $P = 0.56$). Results were consistent if we restricted the data to 303 runs where urbanized sites were tested with a concurrent paired natural site and whether we modelled predation types together or separately (Extended Data Tables 2 and 3 and Extended Data Fig. 1). Urbanized sites in our study had significantly less area with untended vegetation, lower local greenness and smaller connected green areas than natural sites (Fig. 1c and Extended Data Table 1). Thus, our results show that despite significant habitat modification, macroecological patterns can be maintained in urbanized areas.
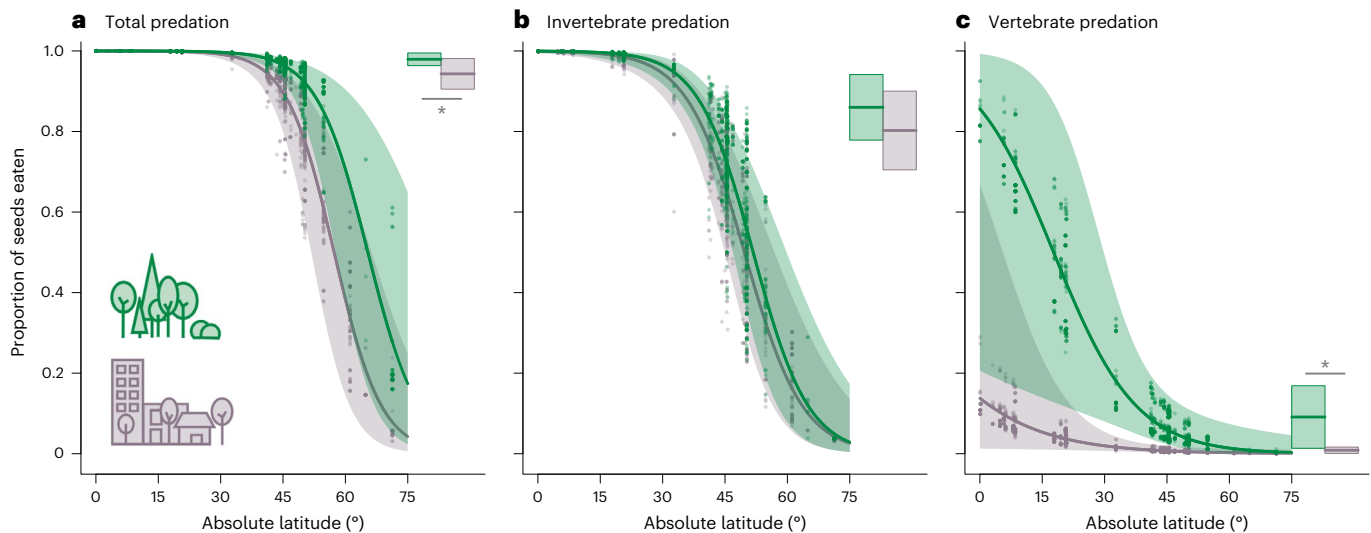
Fig. 2 | Latitudinal gradients in seed predation in natural and urbanized areas. Seed predation declined significantly towards higher latitudes for all predation types. Latitudinal patterns did not vary with urbanization for any predator group, that is, green (higher) and purple (lower) lines do not differ significantly in slope for any panel (the apparently differing slopes in **c** are visually exaggerated due to back transformation from the logit scale). Bars show mean predation intensity (±95% CI) across latitudes (same *y* axis as trend lines); total and vertebrate predation were significantly lower (*) in urbanized (right, purple bars) versus natural areas (left, green bars). Trends, means, 95% CI and partial residuals were extracted from one binomial generalized linear mixed model (GLMM) per predation type to show the independent effects of urbanization on each predator group (results from overall model shown in Extended Data Fig. 1). All results are shown for the median elevation (450 masl). Sample sizes: 156 experimental runs at 36 natural sites and 181 runs at 45 urbanized sites, with 2,912, 2,394 and 2,718 seed depots measuring total (**a**), invertebrate (**b**) and vertebrate (**c**) predation, respectively.

While latitudinal gradients in predation were maintained in urbanized areas, predation intensity was not. Urbanization reduced overall predation on seeds (Fig. 2a), although this effect was not uniform across predators (urbanization × predation type: $x^2_{d.f.=2} = 18.1$, $P = 0.0001$). Urbanization had little effect on predation by invertebrates (Fig. 2b) but significantly reduced predation by vertebrates (Fig. 2c). Changes in the intensity and agents of predation can alter the selective regime experienced by prey and thus the evolutionary trajectory of urban populations[49], as different predator guilds impose differing selection on traits of prey, in this case, seeds[50]. Large-scale effects of urbanization on predator-driven selection have been inferred from differences in the relative abundance of consumers[51] and prevalence of anti-predator/herbivore defenses[16,52] in urban versus natural areas. Our results provide large-scale evidence for a mechanistic link: urbanization changes the relative intensity of seed predation by different predator groups across latitudes.

Altered predation intensity in urbanized areas was not surprising, but the nature of effects was unexpected. One might predict urbanization to reduce predation by invertebrates, contrary to our results, since urbanization can strongly reduce invertebrate diversity and abundance[18,20,53]. In particular, ants were our most commonly detected seed predators (Extended Data Table 4) and urbanization reduces ant diversity, especially in the tropics[30,54]. However, across latitudes we observed ant predation more often in urbanized than in natural areas (Fig. 3). So, either granivorous ants are not suffering the same urbanization-driven declines in diversity as ants in general[30], or their abundance and activity are decoupled from diversity. Indeed, our results suggest that any lost diversity of seed-eating invertebrates in urbanized areas is being made up for by increased abundance or activity of remaining species[55]. This result begs the more general question of how well diversity predicts the intensity of species interactions. Latitudinal gradients in diversity inspired the original hypotheses for latitudinal gradients in interaction intensity[4], but testing this link has been challenging as diversity covaries with other potential drivers of interaction intensity (for example, temperature, productivity[12,13,31]). By disrupting correlations between local diversity, latitude and climate[40], urbanization offers a tantalizing opportunity to disentangle the potential drivers of geographic patterns in predation intensity.
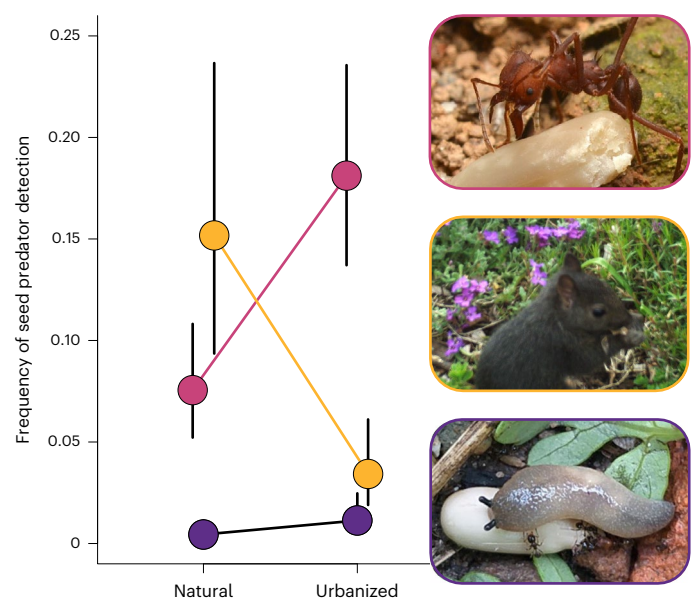
Fig. 3 | Effect of urbanization on observed seed predators. Of the 5,829 seed depots in which seeds were eaten, we were able to identify the seed predators for 1,469 depots. For the three most commonly identified seed predator types (ants, small mammals, molluscs), we tested whether their predation was detected at a higher frequency (that is, at more seed depots) in urbanized vs natural sites. Ant predation was more common in urbanized sites (pink; urbanization $x^2_{d.f.=2} = 12.8$, $P = 0.0003$; $n = 2,394$ caged depots in total), whereas small-mammal predation was more frequently detected in natural sites (light orange: $x^2_{d.f.=2} = 15.3$, $P < 0.0001$; $n = 2,718$ oat depots). Predation by molluscs (snails and slugs) was equally common in urbanized and natural sites (dark purple; $x^2_{d.f.=2} = 2.7$, $P = 0.10$; $n = 8,024$ depots). Points show the mean proportion of depots at which the predator was detected (actual predation rates of all three predators are probably higher), plotted for the median latitude and elevation, ±95% CI. Full statistical details in Extended Data Table 4. Photos from top: leaf cutter ant consuming sunflower seed (Brazil, 21° S; L.P.); eastern grey squirrel eating oats (Canada, 44° N; A.L.H.); slug eating sunflower seed (Mexico, 21° N; C.I.G.-J.).

Fig. 4 | Experimental sites. Urbanized sites (**a**–**e**) often appear more similar to each other than natural sites (**f**–**j**) and often share more similar ground vegetation (**k**–**o**) than natural sites (**p**–**t**). Row **a**: coastal British Columbia (50° N; wet conifer forest biome); row **b**: interior British Columbia (50° N; dry conifer–grassland biome); row **c**: southern Alberta (50° N; prairie biome); row **d**: Great lakes–St Lawrence lowlands, Canada (44° N; mixed hardwood forest biome); row **e**: southeastern Brazil (21° S; tropical rainforest biome). Arrows indicate depots; vertebrate exclusion cages shown in **n** and **s**. Vegetation types: **k**,**m**–**o** = lawn, **l** = garden, **p**–**t** = natural litter. Photo credits: **a**,**f**,**k**,**p** = T.V.; **b**,**g**,**l**,**q** = J.E.; **c**,**h**,**m**,**r** = J.A.L.-Y.; **d** = H.L.S.; **i**,**n** = J.L.; **e**,**j**,**o**,**t** = L.P.; **s** = S.K.-N.

Conversely, given that small mammals and birds can reach high abundances in urbanized areas[56–58], reduced seed predation by vertebrates was surprising. In particular, we detected signs of small-mammal predation significantly less often in urbanized vs natural areas (Fig. 3). It is possible that vertebrate seed predators are still abundant in urbanized areas but seeking higher-reward foods[59] and ignoring experimental seeds. However, it is unclear why richer urban food supply would not also distract invertebrates from experimental seeds and why it would not eventually lead to increased seed predator abundance, resulting in spill-over predation on experimental seeds. Alternatively, our findings could reflect negative impacts of urbanization on the abundance or foraging activity of granivorous birds and mammals[60] across latitudes.

The urbanized and natural sites in our study differed significantly in habitat modification (Fig. 1c), but there was also considerable variation in modification within each category. Urbanized sites ranged from small towns of less than 3,000 inhabitants to cities of more than 4 million, and natural sites varied from small urban parks to extensive wilderness areas of several hundred square kilometres (Fig. 1c). As the ecological impact of urbanization often varies with the amount and type of greenspace remaining[17,20,56], we tested whether more direct measures of local greenness better predicted seed predation intensity. The percentage of greenspace within a 500 m radius of each site and the total area of greenspace connected to the site explained invertebrate predation slightly better than simply classifying a site as urbanized or natural (slight improvements in marginal R^2 for GLMMs and Akaike information criterion (AIC)) but explained vertebrate and total predation slightly worse (Extended Data Table 5). Our core result, that the latitudinal gradients in predation seen in highly natural areas are maintained in more urbanized areas, remained unchanged (that is, latitude × greenness interactions not significant; Extended Data Table 5). Nevertheless, there are many ways to measure urbanization[61,62]. Future experiments specifically designed to test how continuous metrics of urbanization affect predation across latitudes could reveal more subtle effects not captured here.

While local greenness did not predict seed predation across sites, seed predation did vary with local ground cover within urbanized sites (Fig. 4 and Extended Data Table 6). Invertebrate predation was lowest for seeds placed in untended areas with natural vegetation and litter,

whereas predation by vertebrates was lowest on lawns (ground vegetation × predation type: $x^2_{d.f.=4} = 16.2$, $P = 0.0027$), perhaps due to lack of overhead cover from predators[63]. Total seed predation was therefore higher in garden beds than in areas of lawn or natural vegetation litter, which somewhat contrasts the larger-scale pattern of higher total predation in natural sites (Extended Data Table 6). Thus, the factors that determine predation intensity at small scales (within urbanized sites) must differ from those acting at larger scales (between urbanized and natural sites)[64].

Similar to most large-scale experiments on species interactions[2], our study examined one interaction at one trophic level. Seed predation is a particularly important interaction in our context, as it is one of the few specifically named in theory about latitudinal gradients in species interactions[8,9], it has dramatic consequences for plants, which form the backbone of terrestrial ecosystems[65], and urbanization can strongly affect seed fates with rapid evolutionary consequences[36]. How urbanization affects latitudinal gradients in other forms of seed predation (for example, by specialist seed predators), predation at other trophic levels, or gradients in other species interactions are important questions that await further large-scale experiments. Indeed, because urbanization disrupts the ecological gradients hypothesized to generate latitudinal gradients in species interactions, such as primary productivity and species diversity[40,41], further studies comparing interaction intensity between urban and natural areas across large latitudinal gradients could help illuminate the ecological causes of geographic variation in species interactions[2].

Finally, our study highlights the robustness of geographic gradients in seed predation. Compared with the first 'BIG experiment' testing latitudinal and elevational gradients in seed predation in the Americas[13], our current sites vary much more in longitude, hemisphere and the size and connectivity of natural areas, yet we recover equally strong and highly consistent patterns. Across our 36 natural sites, a seed's chance of being eaten increased by 10% for every 10° decline in latitude, remarkably similar to the latitudinal cline found across 79 different sites in different years[13]. Predation by invertebrates in general (Fig. 2b) and ants in particular (Extended Data Table 4) increased strongly from high to low latitudes, whereas latitudinal gradients in vertebrate predation were much shallower, consistent with previous conclusions that invertebrates drive latitudinal gradients in seed predation intensity[13]. While our current study was not designed to test elevational effects, seed predation increased strongly from high to low elevations in all analyses, as found previously (Extended Data Table 3 and Fig. 2). To our knowledge, this is the first time such a large, standardized experiment on interaction strength has been replicated. Our work demonstrates remarkable consistency in continent-wide gradients in seed predation, both across time and space and in the face of extensive habitat modification from urbanization.

Methods

Experimental design

We included sites anywhere on the mainland of the Americas, excluding islands as they experience unique biogeographic processes that might obscure latitudinal patterns. We made sure to have good representation at both tropical (<23.5° absolute latitude) and high (>54°) latitudes, which are often undersampled in both meta-analyses and distributed experiments[2]. In total, we ran the experiment in 8 countries spanning 112° of latitude. We ran the experiment 1 to 11 times per site (median = 4 runs, where a run is one 24 h assay of predation at one site), pairing runs between an urbanized and a nearby natural site within a few days of each other when possible. Replicate runs within a site were separated by at least 2 weeks. Data were collected from 2020 to 2023. While the experiment spanned coronavirus disease 2019 (COVID-19) pandemic-related travel restrictions at some sites, there was no indication that seed predation at our sites changed through time (see 'Statistical analyses' below). We could not fully standardize for site

phenology as tropical and temperate sites differ in seasonality. Rather, we conducted experiments during the snow-free growing season at each site (that is, any time of year in the tropics), during weather typical for that site in that season (for example, avoiding cold snaps, or severe or unseasonal rains).

Each collaborator measured seed predation in an urbanized site and, ideally, a nearby natural area (Fig. 4; some collaborators ran the experiment in additional urbanized or natural sites as well). Urbanized sites were generally backyards, defined loosely as the unpaved (no asphalt, gravel or stonework) and un-built area around a home, be it an apartment building, row house, detached house, or university residence, but in two cases were urban parks or vacant lots. As we only placed seed depots on unpaved ground, urbanized sites had to have some such areas (for example, green balconies, cobblestone courtyards or large parking lots would be ineligible). We used only unpaved ground as seeds that fall on built or paved surfaces generally cannot germinate, making seed predation less ecologically relevant from the plant's perspective.

Urbanized sites were thus unified in that they: (1) were all beside built structures continuously occupied by people (homes but also stores and other buildings in some cases); (2) were close to driveways and roads which separated them from other greenspace; and (3) had their original, native vegetation cleared to make way for homes, replaced with modified vegetation (for example, mowed lawns, garden beds). Some of these sites were highly urban (such as downtown Montreal or Guadalajara, <5% of surrounding area was vegetated), while others were in much greener areas (Fig. 1c). For this reason, we use the term 'urbanized' rather than 'urban' throughout. Paired natural sites had the natural vegetation that would have presumably occupied the urbanized site before vegetation clearing, at roughly the same elevation. Natural sites were generally less than 10 km but up to 40 km from a paired urban site.

Quantifying seed predation

We measured predation on standardized seeds that are widely palatable to diverse seed predators and within the range of natural seed sizes available across our latitudinal gradient[13]. Standardized generic prey that are not local to any site are ideal for measuring cross-ecosystem patterns in predation. Whereas local prey reveal current losses to predation[66], standardized prey control for geographic variation in evolved defenses and phylogenetic history. They reveal the underlying geographic patterns in predation rates that are central to theory on latitudinal gradients in species interactions, which may differ from patterns in realized predation[2,46,67].

This is an extension of the Biotic Interaction Gradients (BIG) experiment. As in previous BIG experiments[13,31], at each site we set out small piles ('depots') of seeds directly on the ground (Fig. 4k–t). We deployed up to 30 depots per site per run as space allowed, separated by at least 6 m (mean of 22.6 depots per run at urbanized sites, 25.3 per run at natural sites). We returned after 24 h to quantify post-dispersal seed predation. To measure underlying predation rates, unclouded by local adaptations between seeds and predators, we used agricultural seeds bred for human consumption; such seeds are not local to any site and should be widely palatable. We only used intact seeds so that damage was unambiguously attributable to granivores. We used sunflower seeds with shells removed (8 seeds per depot; Fig. 4l) and oat seeds with their thin hulls intact (5 seeds per depot, Fig. 4q).

We are familiar with the predators of sunflower and oat seeds from direct observations during depot checks (at 24 h and sometimes earlier checks assessing how quickly seeds were eaten), opportunistic camera-trap observations, and ad hoc feeding and exclusion trials by A.L.H. during this and previous BIG experiments (Supplementary Fig. 1). Oil-based sunflower seeds are eaten by diverse predators, including small mammals (including mice, voles or shrews (detected via faeces), rats (camera traps), squirrels (camera traps)); birds (including

jays, robins, sparrows, finches (camera traps)); and a diverse array of invertebrates including ants, slugs, snails, beetles, earwigs and isopods (seen eating seeds during depot checks; ants and slugs also readily consume sunflower seeds during feeding trials)[13,31]. Sunflower seeds therefore measure total predation by the full suite of seed-predator groups. To measure predation by invertebrates alone, we excluded vertebrates from some sunflower depots using cages of ½" (~1.3 cm) mesh (Fig. 4s).

In contrast, it is difficult to exclude invertebrates from seeds while allowing vertebrates access, and vertebrate predation cannot be calculated by subtracting invertebrate predation from total predation (for example, if invertebrates eat 100% of seeds when vertebrates are excluded, that does not mean that vertebrates would eat 0% of the seeds if they had access). Thus, we assessed vertebrate predation using oats. Carbohydrate-based oat seeds are eaten by small mammals (detected via faeces left in depots) and birds (detected via camera traps). While we did not conduct systematic vertebrate exclusion of oats, observations suggest that oats are generally ignored by invertebrates. Across 5,376 oat depots deployed here and in ref. 13, only 1 of 26,880 oat seeds was eaten by ants (1 ant found eating a seed at 24 h check; other 4 oats intact). Mollusc predation on oats was seen twice and isopod predation seen once. In all cases, invertebrates had made only small holes in oats after 24 h. Predation on oats thus reflects mostly-vertebrate predation.

After 24 h, we searched for seed remains in and around each depot. We counted seeds whose endosperm was partially eaten, fully eaten (but with sunflower seed skins or oat hulls remaining), or removed. We noted signs of predators, for example, rodent faeces, slug slime trails, invertebrates still eating seeds. Seeds removed from the ground can occasionally be dispersed rather than eaten, known as secondary dispersal. Nevertheless, we counted missing seeds as eaten for three reasons: (1) more than 90% of seeds removed from the ground are eaten[33]; (2) our seeds have no adaptations to facilitate secondary dispersal (for example, no thick shells or elaiosomes), making secondary dispersal especially unlikely; and (3) observations and camera-trap footage show that many of our major predator groups remove both sunflower and oat seeds and consume them immediately, suggesting that most removed seeds are eaten rather than dispersed. All birds captured on camera traps consumed seeds immediately. Ants were often seen removing seeds from depots but were almost always breaking seeds up and eating them at the same time.

Identifying seed predators
In most cases of seed predation, we did not find definitive signs of the seed predator (4,356 of 5,825 depots with predation). However, for about a quarter of depots with predation (1,469 of 5,825 depots), we were able to identify seed predators as follows.

Ant predation was noted during depot checks if ants were seen eating seeds (either in depots or while carrying seeds away from depots), if ant hills had been built inside the depot (usually in cages), or if all seeds had been removed but ants were still swarming the depot.

Mollusc predation was noted if snails or slugs were seen eating seeds, or if seeds had been eaten and there were slime trails through the depot.

Mammals were never seen directly, but small mammals (for example, mice, voles) sometimes left faeces in depots (Supplementary Fig. 1f). When faeces were found in oat depots, we often also found the thin hulls of oat seeds left behind (Supplementary Fig. 1f); we therefore considered depots in which oats had been eaten but hulls left behind to be predated by small mammals. If cages were dug up or under, or if seeds had been eaten and the wooden popsicle stick marking the depot was chewed with visible incisor marks, we counted that as signs of mammal predation.

Other predator types (for example, birds caught on camera traps, beetles or other invertebrates seen eating seeds) were recorded but were rare.

These observations are useful for comparison among sites but should be interpreted with nuance. First, ant predation usually needed to be directly observed, so in climates or sites where ants were particularly active and removed all seeds early on, ant predation would be underestimated. Second, predator groups can affect each other. For example, it is only possible to see ant predation if vertebrates have not already eaten seeds and vice versa. When comparing predator observations among sites, we took these constraints into account as follows.

- Models of ant predation considered only caged depots, which measure invertebrate predation rates independent of vertebrate predation. Cages also made it more difficult for ants to completely remove seeds, meaning we were more likely to see ant predation still in action after 24 h.
- Models of mammal predation considered only oats, as (1) oats were rarely eaten by invertebrates and so provide a measure of mammal predation independent of invertebrate predation and (2) mammals often peeled hulls from oat seeds and left them in the depot, so we were better able to detect mammal predation on oats than on sunflower seeds, which were eaten whole.
- Models of mollusc predation considered all depot types, as molluscs often left slime trails so were easily observed in both caged and uncaged depots. We included oat depots, as we have twice seen molluscs eating oats (of >5,825 oat depots in this and other BIG experiments). More often, molluscs left slime trails in oat depots without eating seeds (in which case depots were not recorded as predated). Results did not change if we considered only caged sunflower depots.

Covariates
Urbanized sites varied greatly in how natural they were and how urbanized their neighbourhoods were, and local natural areas also varied in size and connectivity. We therefore investigated covariates that might explain seed predation rates better than our simple categorization of urbanized or natural. First, during experiments we recorded the ground 'vegetation type' surrounding each depot (a microsite): lawn (primarily grass that is regularly mowed); garden (planted and tended areas with plants surrounded by bare soil, mulch, or non-grass ground cover); or untended areas with natural vegetation litter (whether or not the litter was from native plants). At each site, we set depots out to sample each habitat type roughly in proportion to its area (for example, if a backyard had 50% lawn and 50% garden, we set out roughly half our depots in each vegetation type). All depots in natural sites were in areas with untended, natural vegetation litter.

Second, we obtained two continuous measures of urbanization. While there are many possible definitions of urbanization intensity, we wanted measures that were applicable to both natural and urbanized areas (ruling out city size, for example), that varied even among sites that were close together (ruling out rasterized data in which urban and natural sites could be in the same pixel), and that might directly affect seed predator populations. We therefore calculated two metrics by hand, scoring the greenness at each site from satellite images (GoogleEarth) using our local knowledge. We calculated the percentage of green area within a 500 m radius of the centre of each experimental site (ImageJ). We first cropped out water bodies (lakes, large rivers). We then quantified the green area, defined as having vegetated ground cover, including gardens, lawns, fields and natural vegetation and excluding 'grey' areas covered with built structures, paving, stonework or gravel. Areas with green canopy but grey ground covering, such as tree canopies extending over roads, were counted as grey. We also calculated the 'connected green area' (m²) for each site as the total green area that could be accessed without crossing grey areas (ignoring fences). This was done using GoogleEarth's polygon feature. GoogleEarth images were downloaded within 6 months of the final experimental run at each site.

Statistical analyses

Analyses were performed in R (v.4.0.2)[68]. Code and data are publicly archived[69]. Analyses of seed predation (proportion of seeds eaten) used GLMMs with a binomial error distribution and logit link. We had 1 data point per depot, and models included random intercepts for run and site (to account for non-independence of depots set out on the same date at a site ('run') and non-independence of repeated measures at a given site, respectively), and an individual-level random intercept (depot) to resolve overdispersion[70]. We tested the significance of model fixed effects, beginning with higher-order interactions, using likelihood ratio tests comparing the resulting ratios to a Chi-squared distribution. When interactions were significant, we assessed which trends or means differed using estimated marginal means with the Tukey correction for multiple comparisons[71]. For visualization, we extracted trendlines and partial residuals from models[72], and estimated 95% confidence intervals (CI) following ref. [70].

Our main analysis asked whether seed predation varied with latitude for total, invertebrate or vertebrate predation. Our full model had fixed effects latitude × urbanization × predation type + elevation. Latitude is absolute latitude in decimal degrees, urbanization is categorical (urbanized or natural) and predation is categorical, as measured by our three depot types (uncaged sunflower, caged sunflower or oat, measuring total, invertebrate or vertebrate predation, respectively). We did not choose sites to vary systematically in elevation, so we simply controlled for its effect as seed predation varies strongly with elevation[13,73]. Our full data set included 337 experimental runs at 81 sites. We explored the robustness of our results (see Extended Data Table 3 and Fig. 1 for full results). First, we reran the overall model including only runs where an urbanized site was tested within 2 weeks of a nearby natural site (303 runs at 74 sites). Second, we ran one model per predation type with fixed effects latitude × urbanization + elevation.

We next explored the effects of surrounding ground vegetation and greenness. We first tested whether our three measures of local vegetation (proportion of depots in microsites with unkept vegetation with natural litter, % green area within a 500 m radius, total connected green area) varied between urbanized and natural sites or with latitude (full statistical details and results in Extended Data Table 1). Second, for urbanized sites, we tested whether predation differed among vegetation types and whether this varied with latitude or among predation types (full model fixed effects: vegetation type × predation type × latitude; Extended Data Table 6). Finally, we tested whether either measure of greenness explained seed predation better than simply classifying a site as urbanized or natural, by replacing the urbanization term in our main analyses with either greenness measure (scaled). The combined effect of greenness and latitude differed among predation types (%greenness × latitude × predation type: $x^2_{d.f.=2} = 7.41$, $P = 0.025$; connected green area × latitude × predation type: $x^2_{d.f.=2} = 63.39$, $P < 0.0001$), so we then ran one model per predation type with fixed effects greenness × latitude + elevation.

Our experiment was not designed to test whether the COVID pandemic affected predation, and 82% of runs took place after any public-health-related movement restrictions were lifted. Nevertheless, since the pandemic famously altered human movement and therefore potentially affected animal movement and predation, we tested whether predation by any seed predator group changed through time in either urban or natural areas (GLMM fixed effects: urbanization × predation type × month + latitude + elevation, random intercepts for site, run and depot as in our main model; month is an integer counting from first to last month of data collection). While the 3-way interaction was significant ($x^2_{d.f.=2} = 11.9$, $P = 0.003$), there was no significant temporal trend for any predation type in either urbanized or natural sites (that is, 95% confidence intervals of estimated marginal means for all six trends overlapped zero). Temporal trends also did not differ between urbanized and natural sites for any predation type (pairwise comparisons of estimated marginal means).

Reporting summary

Further information on research design is available in the Nature Portfolio Reporting Summary linked to this article.

Data availability

Data used in analyses are publicly available on Borealis, the Canadian Dataverse Repository at https://doi.org/10.5683/SP3/I5DWFQ ref. [69].

Code availability

R code for analyses and figures are also publicly available on Borealis[69].

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Author contributions

Authors are listed in order of contribution. A.L.H. designed and coordinated the study, analysed the data and wrote the manuscript. All authors contributed to fieldwork, except O.R.; O.R. and K.L. developed greenness methods and collected greenness data. E.D.-G. made cages and shipped standardized seeds to collaborators in the USA. O.R., J. Burkiewicz, S.H., M.B.B.-B., E.M., M.L.C., T.L.S., M.C.C., I.J.M.TG., S.K.-N., L.P., S.J., K.M., J.F.B., B.F., J.L. and E.S. helped edit the manuscript.

Competing interests

The authors declare no competing interests.

Additional information

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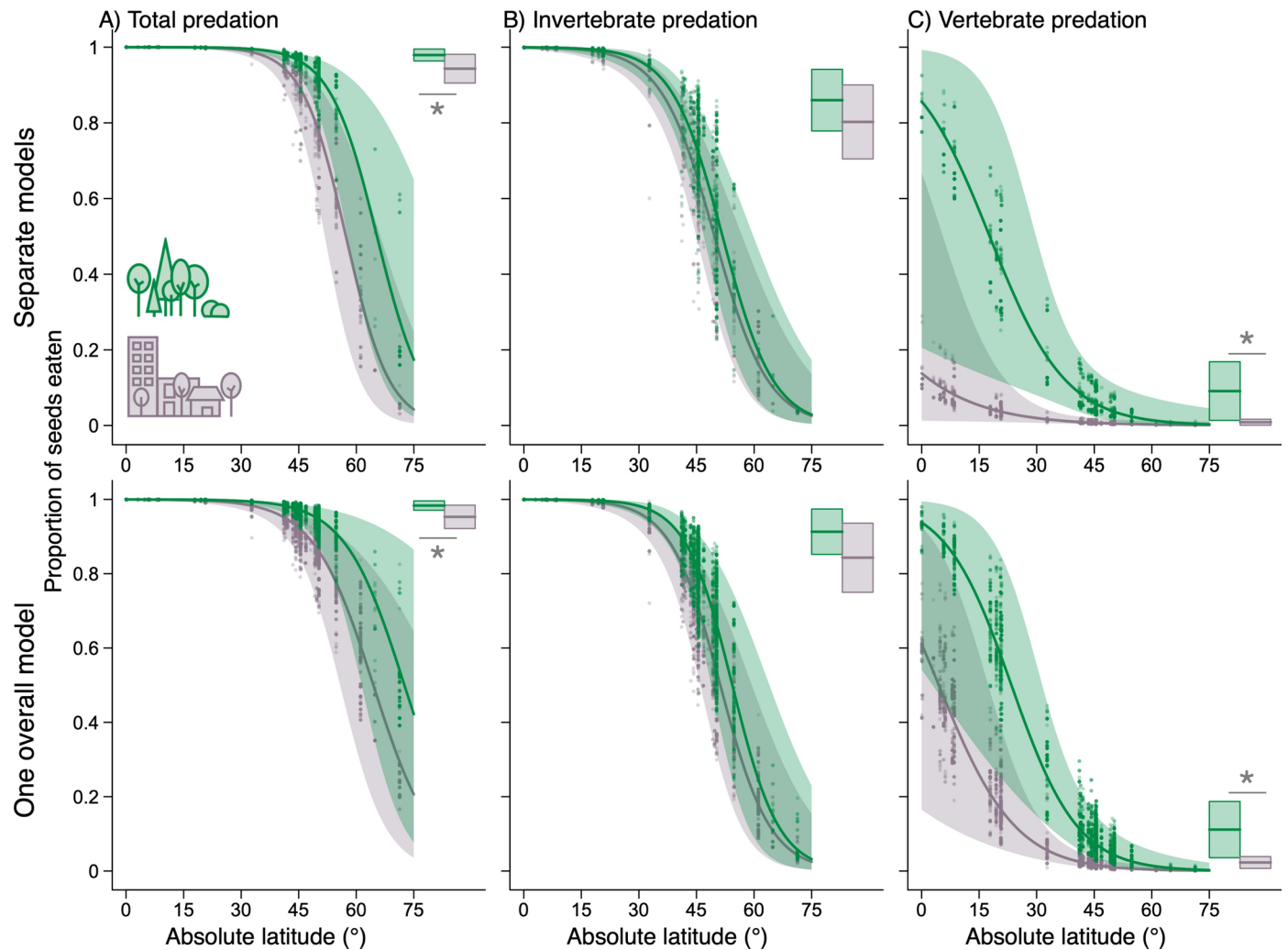
Extended Data Fig. 1 | Separate models per predation types vs. one overall model for all predation types. Top row is the same as Fig. 2, and shows results from 1 model per predation type (column a: total predation by vertebrates and invertebrates, measured using uncaged sunflower seeds; column b: predation by invertebrates only, measured using sunflower seeds caged to exclude vertebrate predators; column c: predation by mostly vertebrates, measured using oat seeds), each with fixed effects Latitude × Urbanization + Elevation. Bottom row shows results from a single overall model including all predation types, with fixed effects Latitude × Urbanization × Predation type + Elevation. Overall conclusions are the same (full statistical results in Extended Data Table 1). Thick lines, shaded polygons and points show means, 95% CI, and partial residuals, respectively. Sample sizes are as in Fig. 2: 156 experimental runs at 36 natural sites and 181 runs at 45 urbanized sites, with 2912, 2394, and 2718 seed depots measuring total, invertebrate, and vertebrate predation, respectively.
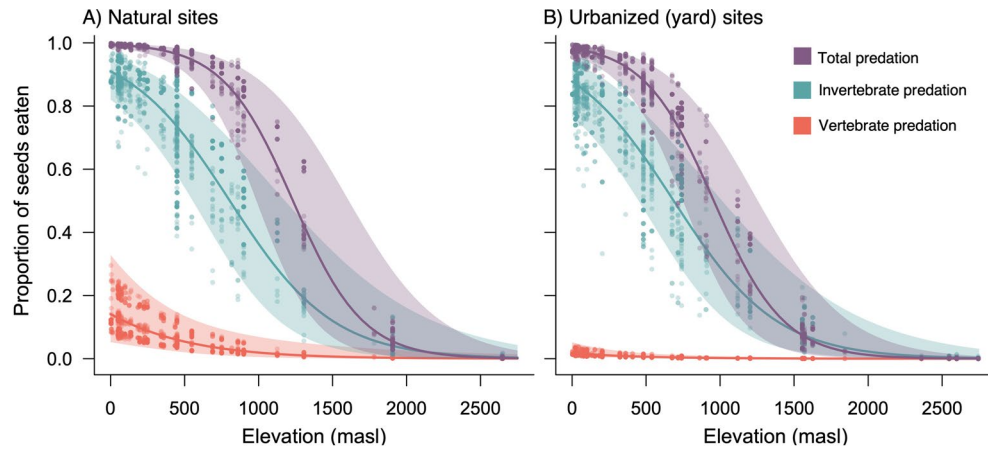
Extended Data Fig. 2 | Seed predation declined toward higher elevations. While our study was not designed to test for elevational patterns in seed predation (unlike[13], which systematically sampled across elevations within each latitude), we included elevation as a covariate in all analyses of seed predation to account for its effects, and it was always significant (Extended Data Table 3). Elevational trends (mean ± 95% CI) and partial residuals are extracted from one binomial GLMM per predation type (fixed effects: Latitude (absolute) × Urbanization (categorical) + Elevation), plotted for the median absolute latitude (45.0°). While we did not test for interactions between elevation and other fixed effects, we have plotted the elevational trends separately for natural (a) and urbanized (b) sites for illustration. Statistical results in Extended Data Table 3.

Extended Data Table 1 | Ground vegetation and local greenness in natural vs. urbanized sites

Response	Response *data structure*	Model type	Latitude x Urbanization df=1	Urbanization df=1	Latitude df=1
depot in untended natural vegetation or not	Binary, *1 data point per depot*	Binomial GLMM	$\chi^2 < 0.001$ $P = 0.997$	$\chi^2 = 114.1$ **$P < 0.00001$**	$\chi^2 = 0.16$ $P = 0.69$
% green area within a 500 m radius	%, *1 data point per site*	Linear	$F = 0.05$ $P = 0.82$	$F = 26.3$ **$P < 0.0001$**	$F = 0.3$ $P = 0.57$
Connected green area (m²)	Log(m²) outliers constrained[1], *1 data point per site*	Linear	$F = 0.46$ $P = 0.50$	$F = 104.7$ **$P < 0.0001$**	$F = 0.1$ $P = 0.78$

[1] Two natural sites backed onto mountainous areas with few through roads dissecting them and so had connected natural areas far exceeding any other sites (140 million m² and 270 million m² whereas the next highest site had a connected green area of 38 million m²). Models did not converge with outliers left in, even after log-transformation. Reasoning that any area greater than 38 million m² was functionally equivalent for seed predators (i.e. very large), we constrained the two outliers to 50 and 60 million m² respectively, enabling models to converge. Constraining the outliers to different values (e.g. both to 50 million m² or both to 38 million m²) produced the same results.

We tested whether each response variable differed between urbanized and natural sites, and whether this difference varied with latitude (model fixed effects = Urbanization (categorical; urbanized or natural), Latitude (absolute), and their interaction). For the proportion of seed depots placed in microsites with natural vegetation and litter, we ran a generalized linear mixed model (GLMM) with a random intercept for site. For greenness analyses, model fixed effects were Urbanization, Latitude, and their interaction. To improve model fit, connected green area was log transformed. Significance of fixed effects was determined using likelihood ratio tests; for GLMMs, GLMs and linear models the resulting ratios are compared to a Chi-squared or *F*-distribution, respectively. Significance of 2-way interaction compares the full model to a model without the interaction. The interaction was never significant so was dropped, and significance of individual terms was determined by comparing a model without the term of interest to the reduced (no interaction) model. A significant effect of Urbanization confirms that sites in urbanized areas differ from those in natural areas. The Latitude × Urbanization interaction tests whether urbanized and natural sites become more similar at one end of the latitudinal gradient, and the Latitude term tests whether the degree of modification varies with latitude, either of which could bias our detection of latitudinal patterns. Results of the Urbanization comparison are plotted in Fig. 1c.

Extended Data Table 2 | Sample sizes

Data included	Site type	Sample sizes	
		Sites	Runs
All runs	Urbanized	45	181
	Natural	36	156
	Total	81	337
Paired runs only	Urbanized	38	148
	Natural	36	155
	Total	74	303

Data are from all runs of the experiment or from only paired runs in which an urbanized site was tested within a couple of weeks of a nearby natural site at a similar elevation and with similar original vegetation. A 'run' is one 24-h assay of predation at one site.

Extended Data Table 3 | Analyses of different data structures yielded consistent results about whether urbanization affects latitudinal patterns in seed predation

A) All predation types in one model

Results of Likelihood Ratio tests (χ^2 statistics) and subsequent Estimated Marginal Means contrasts (z statistics)

Urban × Lat × Pred type	Urban × Lat	Urban × Pred type	Lat × Pred type	Elevation
All runs				
$\chi^2_{df=2} = 1.15$ $P = 0.56$	$\chi^2_{df=1} = 0.11$ $P = 0.74$	$\chi^2_{df=2} = 18.12$ $P = 0.0001$	$\chi^2_{df=2} = 54.27$ $P < 0.0000001$	$\chi^2_{df=1} = 26.69$ $P = 0.0000002$
		Urban. significant?	Latitudinal trend significant?	
		Total: $z = 2.1$; $P = 0.039$	Yes b	$\chi^2_{df=1} = 43.51$ $P < 0.0000001$
		Invert.: $z = 1.3$; $P = 0.20$	Yes a (steepest)	$\chi^2_{df=1} = 25.67$ $P = 0.0000004$
		Vertebrate: $z = 3.2$; $P = 0.0013$	Yes ab	$\chi^2_{df=1} = 9.31$ $P = 0.0023$
Paired runs only				
$\chi^2_{df=2} = 9.18$ $P = 0.010$	not explored further due to significant 3-way interaction, see results from 1 model per predation type below			$\chi^2_{df=1} = 19.84$ $P = 0.000008$

B) One model per predation type

Predation type modelled	Urban × Lat	Urbanization	Latitude	Elevation
All runs				
Total	$\chi^2_{df=1} = 0.19$ $P = 0.66$	$\chi^2 = 4.07$ $P = 0.044$	$\chi^2 = 40.09$ $P < 0.0000001$	$\chi^2_{df=1} = 43.51$ $P < 0.0000001$
Invertebrate	$\chi^2_{df=1} = 0.07$ $P = 0.79$	$\chi^2 = 0.73$ $P = 0.39$	$\chi^2 = 34.28$ $P < 0.0000001$	$\chi^2_{df=1} = 25.67$ $P = 0.0000004$
Vertebrate	$\chi^2_{df=1} = 0.55$ $P = 0.46$	$\chi^2 = 13.63$ $P = 0.00022$	$\chi^2 = 7.77$ $P = 0.0053$	$\chi^2_{df=1} = 9.31$ $P = 0.0023$
Paired runs only				
Total	$\chi^2_{df=1} = 0.84$ $P = 0.36$	$\chi^2 = 1.59$ $P = 0.21$	$\chi^2 = 35.11$ $P < 0.0000001$	$\chi^2_{df=1} = 34.20$ $P < 0.0000001$
Invertebrate	$\chi^2_{df=1} = 0.27$ $P = 0.60$	$\chi^2 = 0.06$ $P = 0.80$	$\chi^2 = 32.49$ $P < 0.0000001$	$\chi^2_{df=1} = 18.37$ $P = 0.00002$
Vertebrate	$\chi^2_{df=1} = 0.999$ $P = 0.999$	$\chi^2 = 13.06$ $P = 0.00030$	$\chi^2 = 7.22$ $P = 0.0072$	$\chi^2_{df=1} = 7.34$ $P = 0.0067$

Results were generally consistent whether we included all predation types in a single model (A) or ran one model per predation type (B), and whether we included data from all runs or only from runs where an urbanized site was tested with a concurrent paired natural site (the only qualitative difference between all and paired runs was in how much urbanization reduced total predation). Sample sizes are in Extended Data Table 2. Likelihood ratio tests compare binomial GLMMs with and without the interaction or fixed effect of interest, using a χ^2 distribution. Fixed effects: Urban = urbanization category (urbanized, natural); Lat = absolute Latitude; Pred type = Predation type (total, invertebrate, vertebrate), Elevation (masl). **A)** When the 3-way interaction (Urban × Lat × Pred type) was not significant it was dropped and significance of 2-way interactions and elevation was then tested against the reduced model without the 3-way interaction. The effect of urbanization and latitude on each predation type was determined using post-hoc tests comparing estimated marginal means (EMM contrasts) from the relevant 2-way interactions, using the Tukey method to correct for multiple comparisons. When the 3-way interaction was significant, we tested the effect of elevation against the full model with the 3-way interaction. **B)** In single-predator models, the effect of elevation was tested against the model with the Urban × Lat interaction. The effects of urbanization and latitude were determined using likelihood ratio tests comparing a model without the interaction or factor of interest to a model with fixed effects Urbanization + Latitude. In all analyses, the Urban × Lat interaction, which tests our main hypothesis that latitudinal gradients in seed predation differ between natural and urbanized areas, was not significant.

Extended Data Table 4 | Seed predator observations varied with urbanization and latitude

Seed predator	Total depots at which predation detected	Depot types included in model (n natural, urbanized)	Results of Likelihood ratio tests			
			Lat × Urbanization (df = 1)	Urbanization (df = 1)	Latitude (df = 1)	Elevation (df=1)
Ants	685	Caged sunflower (1135,1259)	$\chi^2 = 0.52$ $P = 0.47$	$\chi^2 = 12.78$ $P = 0.0003$	$\chi^2 = 4.26$ $P = 0.039$	$\chi^2 = 3.45$ $P = 0.063$
Mammal	538	Oats (1344,1374)	$\chi^2 = 0.33$ $P = 0.57$	$\chi^2 = 15.34$ $P < 0.0001$	$\chi^2 = 0.20$ $P = 0.66$	$\chi^2 = 0.48$ $P = 0.49$
Mollusc	263	All (3938,4086)	$\chi^2 = 0.19$ $P = 0.66$	$\chi^2 = 2.80$ $P = 0.094$	$\chi^2 = 2.58$ $P = 0.11$	$\chi^2 = 0.48$ $P = 0.49$
Other	54	–	–	–	–	–

For our three most commonly detected seed predators, we tested whether they were detected more often in natural or urbanized sites and whether this varied with latitude (one binomial GLMM per predator type). Full model fixed effects were Urbanization × Latitude + Elevation, with a random intercept for site. Likelihood ratio tests first compare models with and without the 2-way interaction; as it was not significant it was dropped and subsequent tests compare models without the fixed effect of interest to the no-interaction model. Ant models consider only predation from caged depots (n = 2394 depots), because caged depots measure invertebrate predation independent of vertebrate predation rates. Further, cages slow seed removal by ants, making it more likely ant predation can still be detected after 24 h. Models of small mammal predation consider only predation on oat depots (n = 2718 depots), as oats are almost entirely ignored by invertebrates and so measure vertebrate predation independent of invertebrate predation. Further, as rodents often strip hulls from oats and leave them in the depot, small mammal predation is easier to detect for oat vs. sunflower seeds. Mollusc presence in depots is easily detected even if all seeds have been eaten, as snails and slugs leave visible slime trails; mollusc models therefore consider all seed types (n = 8024 depots; results are consistent if we consider only caged depots).

Extended Data Table 5 | Relative fit and explanatory power of different measures of greenness

Measure of greenness	Predation type	Greenness × Lat	Greenness	ΔAIC	ΔR²$_{GLMM}$
% greenspace within 500 m					
	Total	$\chi^2 = 0.58$ $P = 0.44$	$\chi^2 = 1.24$ $P = 0.26$	-2.8	-0.0079
	Invertebrate	$\chi^2 = 0.19$ $P = 0.66$	$\chi^2 = 3.62$ $P = 0.057$	3.1	0.0047
	Vertebrate	$\chi^2 = 1.10$ $P = 0.29$	$\chi^2 = 0.04$ $P = 0.85$	-13.6	-0.0341
connected green area (m²)					
	Total	$\chi^2 = 0.91$ $P = 0.34$	$\chi^2 = 0.90$ $P = 0.34$	-3.2	-0.0035
	Invertebrate	**$\chi^2 = 4.09*$** **$P = 0.043$**	–	1.9	0.0361
	Vertebrate	$\chi^2 = 0.76$ $P = 0.38$	$\chi^2 = 0.67$ $P = 0.41$	-13.0	-0.0318

* Interaction remains significant if the two outliers are constrained to the same value or to the third highest value but becomes non-significant if they are removed ($\chi^2 = 2.01$, $P = 0.16$).

All models are binomial GLMMs, 1 per predation type. Full model fixed effects are absolute Latitude × Greenness measure. As described in Extended Data Table 1, the 'connected green area (m²)' predictor includes two outlier sites with extremely large areas of connected greenness (140 million and 270 million m²) which we constrained to 50 and 60 million m² respectively, enabling models to converge. Results are consistent if outliers are both constrained to 50 million m², both to the third highest value (38 million m²), or dropped entirely, except in the case with '*'. Models include random intercepts for run (date within site), site, and observation, except the invertebrate model for % greenspace, in which the random intercept for run is replaced with a simpler random intercept for date (not nested within site) to improve convergence. Significance of terms is determined using likelihood ratio tests (comparing models with fixed effects 'Greenness × Lat' to 'Greenness + Lat', or 'Greenness + Lat' to 'Lat'). Δ values compare the reduced greenness model (that is non-significant Greenness x Lat interaction removed) to the equivalent model (same random effects structure) with categorical Urbanization + Lat as fixed effects (results are the same if we compare full models). Δs calculated such that negative values mean the greenness models perform worse (that is have higher AIC or lower R²$_{GLMM}$): ΔAIC = AIC$_{cat.urban}$ − AIC$_{greenness}$, and ΔR²$_{GLMM}$ = R²$_{greenness}$ − R²$_{cat.urban}$. R²$_{GLMM}$ is the marginal (pseudo) R² for GLMMs, which shows the relative explanatory power of model fixed effects [1]. Binomial variance is calculated as the observation-level variance (delta method in the MuMIn package [2]).

Extended Data Table 6 | Effect of ground vegetation type on seed predation in urbanized sites

	Results of Likelihood ratio tests (LRT)		
Fixed effect of interest	Lat × Pred × Vegetation	Lat × Vegetation	Pred × Vegetation
Models compared in likelihood ratio test (fixed effects only	L×P×V + E L×P + L×V + P×V + E	L×P + L×V + P×V + E L×P + P×V + E	L×P + L×V + P×V + E L×P + L×V + E
Test statistic	$\chi^2_{df=4} = 1.34$	$\chi^2_{df=2} = 4.14$	$\chi^2_{df=4} = \mathbf{16.23}$
Significance	$P = 0.86$	$P = 0.13$	$\mathbf{P = 0.0027}$

Vegetation type is the ground vegetation where each depot was placed; lawn (grass and other herbaceous cover, regularly mowed), garden (planted beds, usually with ground cover of bare soil or mulch), or natural (natural, untended vegetation and litter, whether or not plants are native). Results are from binomial GLMMs. The full model has the fixed effects absolute Latitude (Lat), Predation type (Pred: total, invertebrate, vertebrate), Vegetation type, and Elevation.

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☐	☒	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
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☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	none (data generated in field)
Data analysis	All analyses done in R version 4.0.2 Packages used to generate statistical tests were lme4 v 1.1.27.1, MuMIn v 1.43.17, emmeans v 1.5.1

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Data and code required for all analyses are being made publicly available on the McGill dataverse server (link will be available the next week once submission passes QA QC)

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Reporting on sex and gender	NA
Reporting on race, ethnicity, or other socially relevant groupings	NA
Population characteristics	NA
Recruitment	NA
Ethics oversight	NA

Note that full information on the approval of the study protocol must also be provided in the manuscript.

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Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

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Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	in all analyses of seed predation, sample size is the number of experimental seed depots of a given depot type (as specified in manuscript). for comparisons of site characteristics, sample size is the number of sites in each category : urban or natural
Data exclusions	no data were excluded except for analyses of the vegetation type each depot was placed in: in this case one run was missing this information and so had to be excluded
Replication	experiments were run multiple times at most site to capture the variation within latitudes and urbanization categories
Randomization	none
Blinding	none

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

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☒	☐ Clinical data
☒	☐ Dual use research of concern
☐	☒ Plants

Methods

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Does the work involve any of these experiments of concern:

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☒ ☐ Demonstrate how to render a vaccine ineffective

☒ ☐ Confer resistance to therapeutically useful antibiotics or antiviral agents

☒ ☐ Enhance the virulence of a pathogen or render a nonpathogen virulent

☒ ☐ Increase transmissibility of a pathogen

☒ ☐ Alter the host range of a pathogen

☒ ☐ Enable evasion of diagnostic/detection modalities

☒ ☐ Enable the weaponization of a biological agent or toxin

☒ ☐ Any other potentially harmful combination of experiments and agents

Plants

Seed stocks	Only standardized experimental seeds were used, purchased from local food stores. For all sites in Canada we used oats purchased from WestCoast seeds, and sunflower seeds of Milanaise brand. For all sites in the USA, we used oats purchased from a horse food supply store. Collaborators in other countries purchased seeds locally.
Novel plant genotypes	NA
Authentication	NA