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Abstract

Urbanization is often associated with homogenization, including the homogenization of biodiversity and overpopulation by generalist species that can change community and disease dynamics. In Madrid, nestlings of Eurasian Scops Owl (*Otus scops*) frequently suffer Necrotic Oropharyngeal Disease as a result of infection by *Gongylonema* sp., a parasitic nematode transmitted by adult owls to their offspring through the diet, more specifically through consumption of a pest species: the oriental cockroach (*Blatta orientalis*). We studied how the presence of cockroaches and owl infection rates are affected by urban features such as green areas. We found that a higher number of birds were affected by the parasite in the initial period of the breeding season and in areas with greater surface area of green spaces. We suggest that urban management should promote the diversity of insects in green areas so that the owls diversify their dietary offer to their offspring, to reduce the prevalence of the disease and improve their breeding success in the city of Madrid, as this problem has not yet been described in other cities.

Key words: urban ecology, disease ecology, pathogens, vector management, urban green area management.

Introduction

Large metropolitan areas are currently undergoing rapid growth associated with numerous negative effects on species richness which may be caused by pollution, habitat fragmentation, loss of primary habitats or loss of resources (Palomino and Carrascal, 2006). Although living in a city can have positive effects for different species (Aronson Mila et al. 2014; Rébolo-Ifrán et al. 2017; Seitz et al. 2022), urbanization is now considered one of the main drivers of biodiversity change, which may cause species loss and homogenization (McKinney, 2006; Pauchard et al., 2006). Studies focusing on the direct or indirect effects of urbanization on species are particularly necessary to detect selective pressures on populations in large urban areas and to assess their adaptation (Fernández-Juricic and Tellería, 2000; Fernández-Juricic et al., 2001). In this context, we have selected the Eurasian Scops Owl (*Otus scops* Linnaeus, 1758), hereafter referred to as Scops Owl, as a model species in the city of Madrid. In Spain, this species suffered a population decline of 32.4 % between 2006 and 2018, and is listed as vulnerable (SEO/BirdLife, 2021). In Madrid, Scops Owl chicks infected with undescribed species of *Gongylonema* (Nematoda) can develop a disease called Necrotic Oropharyngeal Disease (NOD; Esperón et al., 2013). Since 1997, 90% of nestlings and fledglings have annually shown severe proliferative necrotic lesions in their oral cavities, which can lead to death by starvation (Lopes et al., 2022). Also, it has only been detected in chicks less than 3 weeks of age. So, it may be associated with reduced productivity and a significant decline of Scops Owls in Madrid (authors' unpublished data). This disease has not been detected in owls in other large cities.

Gongylonema has indirect life cycles associated with coprophagous arthropods, such as dung beetles or cockroaches, which act as intermediate hosts (Illescas-Gómez et al., 1988; Jelinek and Löscher, 1994). The oriental cockroach (*Blatta orientalis* Linnaeus, 1758, Arthropoda: Blattodea) has recently been identified as the intermediary host for the *Gongylonema* sp. of Scops Owl in Madrid. Almost 67% of analysed cockroaches in this city were infected, and cockroaches are the most common prey that adults provide to the chicks in this area (Lopes et al., 2022). In Madrid, this cockroach is considered a common and annual pest, even causing public health concerns (Madrid Salud, 2016). In general, arthropods are negatively impacted by urbanization, which may cause a drastic decrease in species richness (McIntyre, 2000; Lagucki et al., 2017; Fenoglio et al., 2020), and arthropod communities

dominated by a few generalist and opportunistic species, such as cockroaches (Penone et al., 2012).

Because of the potential impact that NOD could be having on the Scops Owl population in Madrid, the aim of this study is to evaluate the environmental factors that determine chick infection by *Gongylonema* sp. Since the oriental cockroach is the only known vector for *Gongylonema* sp., it is hypothesized that a higher NOD prevalence will be temporally and spatially associated with these insects, such that a higher incidence of infection in Scops Owls during their breeding season would be expected. These data will be critical to identify and implement appropriate management activities in the cities.

Material and methods

Study area

The study was carried out in the city of Madrid (Figure 1), the most populated city in Spain, with over 3.2 million inhabitants (National Statistics Institute, 2018). Madrid is highly urbanized with an extensive network of buildings, sanitation and sewage systems, but has a high density of green spaces representing 9% of the municipality's surface area (according to data from the latest Municipal Barometer of Urban Economy of the Madrid City Council).

Data collection

Between 1997 to 2015, data were collected from all Scops Owls admitted to the Brinzal Night Raptors Recovery Center. For each case, the presence or absence of NOD, the georeferenced location where the individual was found and the admission date to the Center (ADC) was recorded. The breeding season was classified from June to September (Mikkola, 1983; Alonso et al., 2003). As the lesions caused by the parasite were only detected in 2-3 week old chicks, data from adult individuals were discarded (Figure 1A).

Variable selection

Although Scops Owl chicks of this age are mobile, they cannot fly, and so it is assumed that the location in which they were found is near their nest. To represent the feeding area of each breeding pair, a 150 m buffer was established. This was the maximum distance that parents move away from the nest to hunt, based on movement of individuals tracked with radio-transmitters (Lopes et al. 2022).

Areas with higher presence of oriental cockroaches in Madrid were identified from the year 2010 to 2015, using reports of cockroaches sights in public areas, provided by the Technical Unit of Vector Control of the Department of Environmental Health of Madrid

(Madrid Salud). Only reports between June and November were selected, representing the highest interaction among the phenology of cockroaches and the Scops Owl chick feeding period. A total of 6,800 oriental cockroach presence reports were provided (Figure 1B).

To understand the factors that determine the presence of cockroaches, a series of variables were selected considering the biology and habitat requirements of the oriental cockroach. These variables included seasonal humidity (average monthly relative humidity in % also related with precipitation variables); minimum temperature (average monthly minimum temperature in °C) and distance to the nearest sewer. To correct possible bias of higher presence of cockroach in the vicinity of the sewage system, the Sewage Unit of the City of Madrid provided a map with a total of 14,568 gratings and registration lids throughout the study area. In addition to such traits, cockroaches are expected to favour areas near rivers, and consequently, the distance to the nearest watercourse was considered.

Statistical analysis

To determine if NOD prevalence among administered owls varied among years, we constructed a General Linear Model (GLM) including year as a covariate. Also, an additional ANOVA test to compare between the observation dates of infected and non-infected chicks throughout the year was performed.

The progression of the number of cockroach reports over the years was analysed with a univariate linear regression. To consider the biology and habitat preferences of the cockroaches, we assessed the variation in the number of cockroach presence alerts in relation to the date (during the breeding season of the owls), humidity, minimum temperature and distance to the sewers with regressions.

To predict the environmental variables that influence NOD presence in Scops Owls, we constructed a binomial GLM with the presence/absence of lesions as a binomial dependent variable. The time of the year in which the chicks were reported as ADC (including the quadratic term), the proportion of green areas (% green area) and the number of sewers in each home range (distance to sewer), humidity, minimum and maximum monthly temperature and distance to the river were used as exploratory variables.

Initially, a preliminary study of all the variables was carried out through an array of correlations. We simplified this model by selection of variables based on the Akaike information criterion (AIC).

All statistical analyses were performed in the R software environment (R Core Team 2022) with the *lme4* (Bates et al. 2015) and *multcomp* packages (Hothorn et al. 2008). Graphs

were produced with the *ggplot2* package (Wickham 2016) while maps and GIS analyses were made with ArcGIS 10.8.

Results

From 1997 to 2015, out of the 477 owl chicks under study, 286 (60%) showed necrotic plaques in their oral cavities consistent with NOD. The distribution of admitted chicks and disease status is shown in Figure 1A.

The number of owls administered increased between 1997 and 2015 (Wald = 5.954; $p < 0.001$) (Figure 2A). However, NOD prevalence has not fluctuated significantly among years (Wald = 19.705; $p = 0.35$) (Figure 2A) and only two years differed significantly from the rest; the highest prevalence in 1998 (Wald = 3.95, $p = 0.047$) and the lowest in 2013 (Wald = 10.51, $p = 0.001$). Thus, data from all years were combined for the rest of the analyses.

There was a significant difference in the date of admission of infected and non-infected chicks in each year ($F_{1, 475} = 14.091$, $p = 0.0002$) (Figure 2B), with infected owls found earlier (mean Julian day = 190.06 = July 9th; SD = 17.15) than non-infected ones (mean Julian day = 197.62 = July 16th; SD = 26.87). Cases of NOD are more frequent early in the season and in areas with a higher percentage cover of green areas (Table 2), while the number of sewers within each breeding pair home range, humidity, maximum temperature and proximity to the river did not have a significant effect on NOD infections. The number of cockroach reports declined significantly between 2010 and 2015 ($r^2 = 0.82$, $p = 0.013$, Fig S1). The regressions revealed a negative relationship between the time of the year and the number of cockroaches (i.e.: within the Scops Owl breeding season, the earlier the date, the more cockroaches there will be and vice versa), as well as with humidity and distance to the sewers, and a positive relationship with temperatures (i.e.: the higher the temperature, the more cockroaches) (Table 1). Additionally, cockroaches were detected more frequently in the green areas closer to sewers ($r^2 = 0.852$, $p = 0.009$).

We also analyzed the frequencies of cockroach occurrence throughout the breeding season of the Scops Owl (June to November) to determine the period in which the highest number of oriental cockroaches occurs. Cockroach activity peaked from mid-June to mid-July (Julian day 160-200) and gradually decreased throughout the rest of the breeding season, which is partially in line with the peak in NOD prevalence in the Scops Owls (Figure 2C), with a difference of a few days (Table S1).

Discussion

This study highlights the importance of urbanization processes in biological communities involving not only predators (*Otus scops*) but also their prey (*Blatta orientalis*). Oriental cockroaches are known to be the main prey of Scops Owls breeding in the urban environment of Madrid, and act as vectors of *Gongylonema* sp. causing lethal infections in Scops Owl chicks. Here we studied the seasonal and long-term dynamics and environmental correlates of *Gongylonema* infections in Scops Owl chicks. The results show that the breeding period of the Scops Owl in the city of Madrid has a temporal relationship with the peak of cockroach abundance, which increases the risk of infection of chicks by *Gongylonema* sp.

We found that in the city of Madrid, cockroaches can be found in greater numbers at the beginning of summer (Figure 2C), when the minimum temperatures are higher (Robinson, 2005). Under natural conditions, Scops Owls show a very diverse insectivorous diet, dominated by large Orthoptera and nocturnal Lepidoptera (Bavoux et al., 1993; Mori et al., 2016). The diet of this species in Madrid seems to differ from that of their non-urban conspecifics, by preying frequently on cockroaches. Lopes et al. (2022) describe cockroaches in Madrid's parks as the most abundant prey, representing 51% of the potential prey for the Scops Owl, and being one of the few available prey species with nocturnal habits. This issue makes cockroaches abundant, conspicuous, and available. In urban ecosystems, parks that represent islands of natural habitat immersed in a large urban matrix (Fernández-Juricic and Jokimäki, 2001), become essential elements for the maintenance of biodiversity in cities (Savard et al., 2000). The problem arises when these green spaces are landscaped and the environmental management strategies for parks are not adequate (i.e: biocides), resulting in limited arthropod diversity (Penone et al., 2012).

Our results showed a highly significant spatial influence related to green areas within Madrid, since more owl individuals were infected with *Gongylonema* sp. in locations with a higher proportion of green spaces within the home range of the breeders (Table 2). Scops Owls in the city use green areas as nesting and breeding sites (Lopes et al., 2022). Oriental cockroach reports (see Figure 1B) mainly refer to strictly urban areas, yet green areas such as city parks in Madrid show very high cockroach abundance and density (Lopes et al., 2022). The Scops Owls use urban habitats with abundant food, but at risk of parasites (Delibes et al., 2001). Therefore, in the urban context of Madrid, changes in habitat management should be promoted to counteract the detrimental effects on biodiversity (Sattler et al., 2010). In particular, by managing parks and green areas to maintain a high diversity of arthropods, exploitation of cockroaches by the Scops Owl and transmission of the parasite could potentially be reduced.

The NOD prevalence in chicks from Madrid has remained nearly constant during this 19-year study period and, given that cockroaches act as a vector, a higher incidence of the infection in the Scops Owl would be expected during the breeding season. In fact, our results show more owl individuals infected with *Gongylonema* sp. at the beginning of the breeding season, around the first half of July (Figure 2B), which partially overlaps with the phenology of the cockroach. The non-complete overlap could be due to the delayed onset of symptoms in the chicks after parental feeding. In addition, our results show a significant relationship between the proximity to the peak of cockroach abundance and the number of chicks infected with *Gongylonema* sp., due to a higher intake of infected oriental cockroaches as the most available resource at the beginning of the season. Consequently, if owls schedule their breeding season to coincide with the availability of cockroaches this could lead to a higher rate of NOD prevalence in Madrid.

Among the possible hypotheses concerning *Gongylonema* life cycle, we propose that Scops Owls are dead-end accidental hosts involved in a vectorial cycle of the *Gongylonema* life cycle, in which cockroaches are vectors and another vertebrate species is the definitive host. Due to high abundance of cockroaches in urban areas of Madrid (Lopes et al., 2022), Scops Owls are forced to feed their chicks with infected cockroaches, resulting in *inter-animal zoonosis*, without affecting the adult owls. Considering that there must be a definitive host with an abundant population settled in the city to complete the life cycle of *Gongylonema* sp., it is most likely that an urban mammal (*Rattus* sp.) can play this role (Setsuda et al., 2016). This cycle would be closed through the organic residues of the definitive hosts, whose carcasses would be ingested by the cockroaches during their feeding activities.

This study highlights the cost of living in urban areas and reaffirms the importance of developed maintenance programs in urban green spaces to increase arthropod richness and feeding resources for Scops Owls, as well as adequate cleaning services and garbage management to control pest species. Higher prey diversity enhances the dilution effect and reduces infection risks (Johnson & Thielges, 2010). Since the parasitic cycle is still unknown, studies should be conducted to detail its development and transmission to hosts.

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Data Availability Statement

Data available on request from the authors: The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Tables and figures

Table 1. Results of regression analyses of the influence of abiotic variables (distance to sewers, relative humidity, time of year and minimum temperature) on the presence of cockroach reports (*Blatta orientalis*) in the city of Madrid. * p<0.05; ** p<0.01; *** p<0.001.

| Variable | Estimate | Std. Error | t value | Pr(> t) | R ² |
|-----------------------------------|----------|------------|---------|------------|----------------|
| Distance to nearest sewer (meter) | -2.112 | 0.615 | -3.433 | 0.0007*** | 0.063 |
| Monthly relative humidity (%) | -1.854 | 0.189 | -9.789 | <0.0001*** | 0.353 |
| Date (Julian day) | -0.641 | 0.028 | -23.160 | <0.0001*** | 0.753 |
| Minimum temperature (°C) | 4.132 | 0.545 | 7.575 | <0.0001*** | 0.246 |

Table 2. Results of final GLM model of the environmental variables influencing NOD presence in Scops Owls in the city of Madrid. The distance to river is the distance in metres between the Scops Owl occurrence and the nearest river, admission date to the centre is the Julian day when the Scops Owls were found and admitted to Brinzal Night Raptor Recovery Center, ADC ^2 is quadratic Julian day, the % green area is the percentage of green and vegetated areas around the Scops Owl occurrence. * p<0.05; ** p<0.01; *** p<0.001.

| Variable | Estimate | Std. Error | z-value | Pr(> z) |
|-----------------------|------------|------------|---------|--------------|
| Distance to river (m) | -7.143E-05 | 4.319e-05 | 1.654 | 0.09813 |
| ADC (julian day) | 0.1757 | 0.06.799 | 2.585 | 0.00974 ** |
| ADC^2 | -4.597e-04 | 1.696e-04 | -2.711 | 0.00671 ** |
| % green area (%) | 0.021251 | 0.003674 | 4.321 | 1.56E-05 *** |

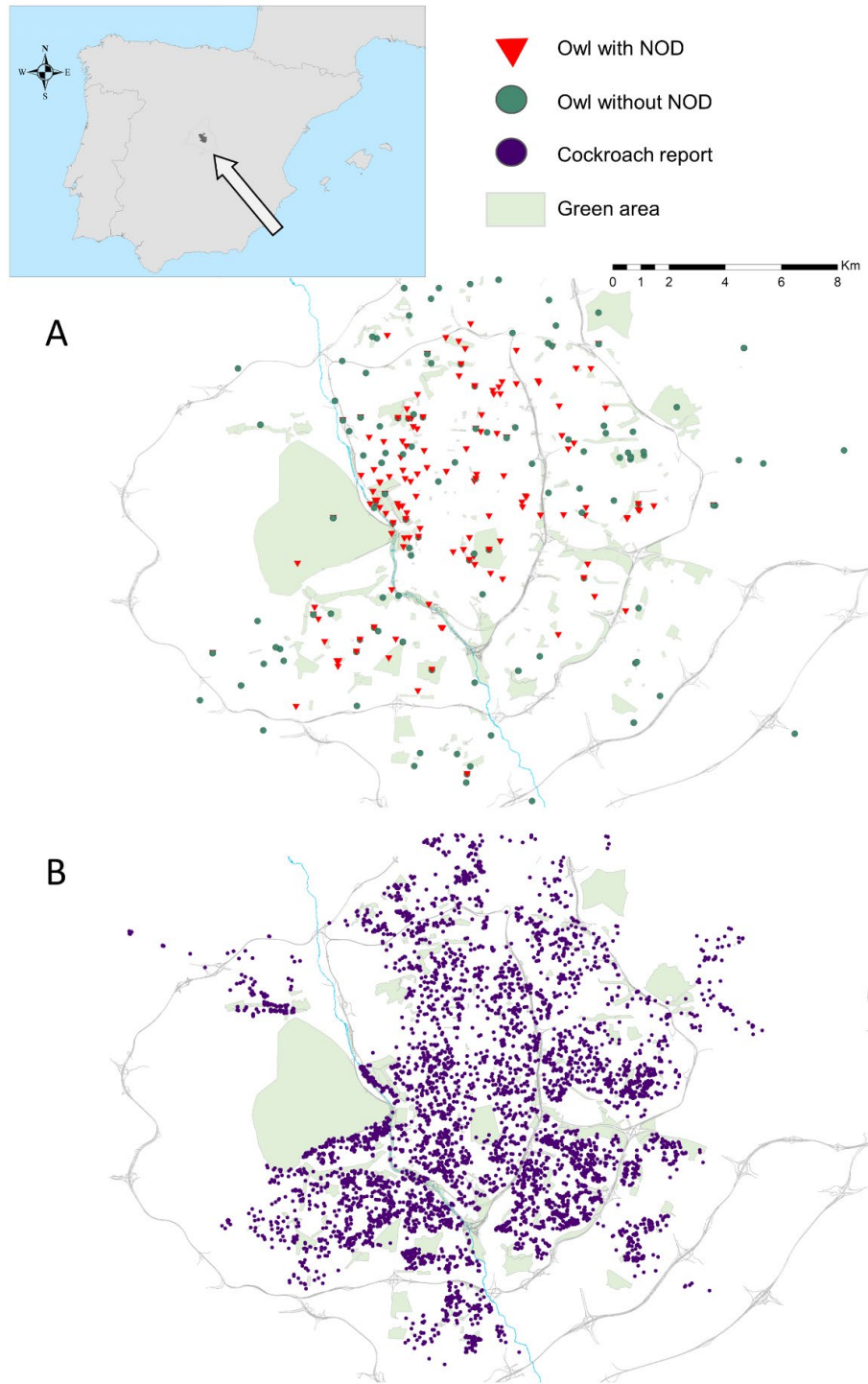


Figure 1. Locations of **A.** Scops Owl chicks infected and not infected with NOD in the city of Madrid (Spain) from 1997 to 2015, and **B.** oriental cockroach reports in the city of Madrid from 2010 to 2015. The blue line indicates the Manzanares River and grey lines indicate the city's ring roads, marking the urban center in the middle of them.

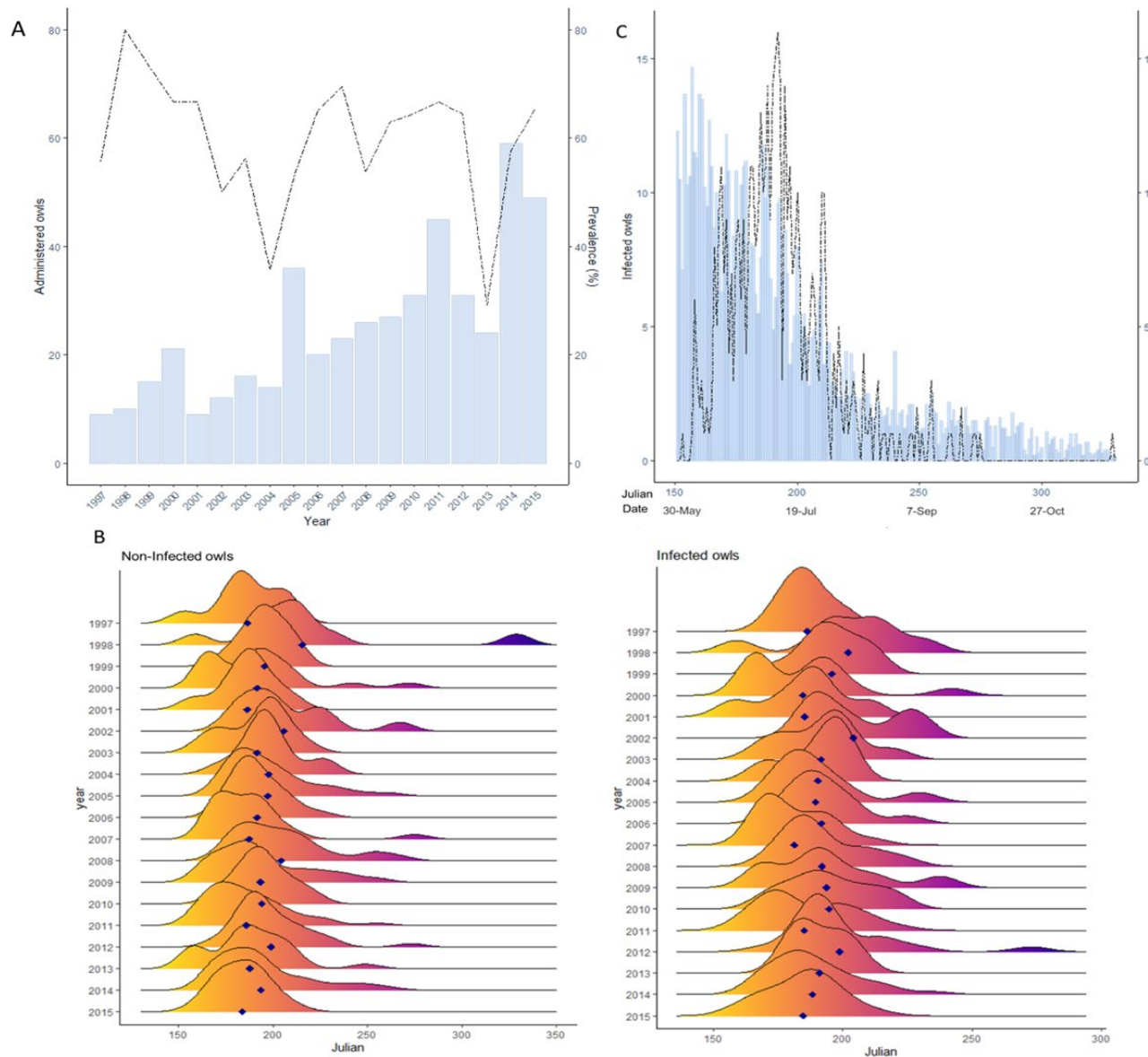


Figure 2. Prevalence of necrotic oropharyngeal disease in Scops Owls from Madrid, Spain, from 1997-2015. **A.** Annual variation in number of owl chicks admitted to the centre (blue bars) and the percentage infected with NOD (dashed line). **B.** Daily number of non-infected (left) and infected (right) owl chicks in each year from 2010-2015. **C.** Daily number (pooled data from 2010 to 2015) of oriental cockroach (*Blatta orientalis*) reports during the Scops Owl breeding season (blue bars) and owl chicks with NOD throughout the year from 1997 to 2015 (black line dashed). Days of the year are shown in the Julian calendar.