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# Spatiotemporal variation in the adult sex ratio, male aggregation, and movement of two tropical cloud forest dung beetles

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## Abstract

While theory suggests that at conception the sex ratio should be balanced (1:1), this can be variable across space and time in wild populations. Currently, studies of the environmental factors that regulate adult sex ratio (ASR) in species with different life history traits are scarce. Using capture–recapture over a year, we analyzed the influence of habitat type (forest and nonforest) and season (rainy and dry) on variation in ASR, male aggregation and the trajectory movement of 2 dung beetle species with different life history traits: *Deltochilum mexicanum* (a hornless roller species) and *Dichotomius satanas* (a tunneler species with horns on its head and thorax). We found opposite tendencies. The *D. mexicanum* population tends to be female-biased, but the population of *D. satanas* tends to be predominantly male, and observed values were not related to habitat type or season. However, the 95% confidence intervals estimated were highly variable between seasons depending on habitat. On examining the monthly variation in ASR for both habitats, we found that it depends on the species. In addition, male aggregation differed between species depending on habitat type and season, and species movement patterns were closely related to their habitat preferences. Based on our results, we argue that comparative population studies of species with different life history traits are necessary to understand the variation in demographic parameters as well as its ecological and evolutionary implications in the face of spatial and climatic environmental variation.

**Key words:** habitat preference, mark-recapture, mean crowding, seasonality, Scarabaeinae, sex proportion.

As proposed by Fisher (1930), natural selection favors a 1:1 sex ratio at conception. However, as repeatedly found for many taxa, the sex ratio tends to be biased toward one of the sexes depending on environmental conditions and the species' life history traits (e.g., mating system, sexual dimorphism, and dispersal capacity; Dittmar et al. 2011; Jennions and Fromhage 2017). Among sex ratio measures, adult sex ratio (ASR), estimated as the proportion of males to females for individuals that have reached reproductive age, is a population parameter related to morphological, physiological, and

behavioral differences between the sexes (Ancona et al. 2017; Jennions and Fromhage 2017). ASR is fundamental to understanding demographic processes, sexual selection, and social evolution (Eberhart-Phillips et al. 2018). Sex ratio biases and their variation may reflect adaptations related to changes in the distribution of individuals due to climate variation, spatiotemporal resource limitations, and parasitic infections, among other factors (Dyson and Hurst 2004; Dittmar et al. 2011; Székely et al. 2014; Jennions and Fromhage 2017; László et al. 2018). ASR bias affects the spatial and

temporal aggregation of organisms, as well as growth rates and the evolutionary trajectories of wild populations (Gownaris et al. 2020).

Spatiotemporal aggregation is common in several taxa and generally occurs when individuals compete intensely for resources that have a patchy small-scale distribution and are ephemeral (Inouye 2005; Pomfret and Knell 2008; Casalini et al. 2010). Both the habitat characteristics and temporal availability of resources such as mates, nesting sites, and food have a differential effect on males and females. These factors influence sex ratio and population density, as well as the time individuals stay in a population, their reproductive fitness, and survival (Julliard 2000; Reichard et al. 2014). Owing to species habitat preferences, it is expected that optimal environments—that is, those that offer suitable conditions for survival such as microclimate, resources, and protection from predators and parasites—would favor high density or aggregation, and sex ratio would be biased toward the sex with the lowest dispersal capacity and thus the lowest probability of death (Julliard 2000; Steifetten and Dale 2012; Végvári et al. 2018). Species' dispersal differentiated by sex is context-specific and has been related to the type and quality of habitat, and is known to influence sex ratio and aggregation (Bell 1990; Kral-O'Brien and Harmon 2021). However, empirical studies on the movement of the sexes and its influence on the structure and demographic processes of populations under natural conditions are scarce (Kral-O'Brien and Harmon 2021).

Variation in the ASR and aggregation has also been related to sex differences in body size and longevity, time of appearance, reproductive rate, and the arrival of the individuals at breeding sites, as well as by the influence of environmental conditions (Julliard 2000; Dittmar et al. 2011; Székely et al. 2014; Carmona-Isunza et al. 2017). In habitats with marked seasonality, it is expected that variations in ASR and aggregation would be correlated with changes in the climate conditions that occur between seasons (Donald 2007; Reichard et al. 2014; Székely et al. 2014). This is important, because in insects, as in other ectothermic organisms, temperature and precipitation are ecophysiological determinants of the variation in ASR, because they influence life cycles through mechanisms such as development and reproduction rate, and often act as a signal for migration and dispersal (Tauber et al. 1998).

Dung beetles of the Scarabaeinae subfamily (Coleoptera: Scarabaeidae) are a model group of insects for analyzing spatiotemporal variation in sex ratio and its relationship to aggregation (Pomfret and Knell 2008). These beetles use ephemeral, highly localized resources, preferentially mammal dung and/or carrion, which serve as a substrate for reproduction and as food for both larvae and adults (Halffter and Edmonds 1982; Bogoni and Hernández 2014). The high degree of competition for these resources has led to diversification in size and morphological structures, and also in their strategies for resource relocation as well as variation in nesting behavior and parental care by 1 or both sexes (Halffter and Edmonds 1982; House et al. 2011). Studies done to date on the sex ratio of dung beetles have focused on the operational sex ratio (OSR, proportion of sexually active adults), and have demonstrated that male-biased OSR and high aggregation predict the sexually selected traits (Pomfret and Knell 2008). It is also known that OSR and density vary among populations that are geographically separated (Moczek 2003) and according to House et al. (2011) in the dung beetle *Onthophagus taurus*, the sex ratio depends on the differential mortality rate between sexes, as a function of the amount of food provided by the parents during nesting. In some dung beetle species (e.g., *Dichotomius satanas*) with an annual reproductive cycle, it is common to find sexually immature individuals during much of the

year (Barretto et al. 2021). This makes ASR a useful estimator, combined with the fact that the sexual maturation of individuals based on external morphological characteristics (e.g., rigid exoskeleton) is difficult to determine (Barretto et al. 2021).

We sampled over the course of a year, and analyzed the influence of habitats with contrasting vegetation cover (forest and nonforest [NF]) and seasonality (rainy and dry) on variation in ASR, male aggregation, and the movement of 2 dung beetles: *Deltochilum mexicanum* (Burmeister, 1848) and *D. satanas* (Harold, 1867). Both species are typical of the cloud forest in Mexico and are abundant most of the year, but differ in habitat preference: *D. mexicanum* almost exclusively inhabits well-conserved cloud forests (Moctezuma et al. 2021), while *D. satanas* is a tolerant species associated with different types of land use, including forest, pastures, and crop fields (Halffter et al. 2007; Rös et al. 2012). They also differ in their expression of secondary sexual characters, dung relocation behavior, nesting pattern, and parental investment (Table 1). Previous studies (Barretto et al. 2018) indicate that, even though population size and survivorship rates are similar, these species differ in recruitment probability (i.e., new individuals in the population), suggesting differences in birth rates and migration, demographic processes that are determinants of population dynamics (Table 1). Based on the species' life history traits, and specifically on differences in secondary sexual traits (Table 1), we hypothesize there will be opposite patterns in the spatial-temporal variation of ASR and different male aggregation patterns. For *D. mexicanum*, a hornless species, we expect ASR to be female-biased. For *D. satanas*, a species with horns on its head and thorax, we expect ASR to be male-biased. Because environmental conditions differentially influence the abundance of each sex, we also expect ASR and male aggregation to be affected by habitat type, and regarding seasonality, we expect a greater aggregation of male for both species in the rainy season, when adult emergence and reproduction occur (Halffter and Matthews 1966; Cambefort and Hanski, 1991).

## Materials and Methods

### Study area

The study was done in a cloud forest landscape between 1,520 and 1,820 elevation in metres above sea level (m a.s.l.), west of the city of Xalapa (19° 30' 55.81" N; 97° 0' 19.88" W) in the municipality of Rancho Viejo, Veracruz, Mexico (Figure 1). The climate of the region is temperate-humid, with 1,500–2,000 mm of rainfall per year. Mean annual temperature is 17–19°C and there are 2 clearly defined seasons: the rainy season from June to October, and the dry season from November to May (Williams-Linera 2002).

### Sampling design and beetle capture

To ensure the capture of both species, we delimited a plot of 126 ha and used the habitat categorization proposed by Barretto et al. (2018): (1) Forest (F), which corresponds to native vegetation (~50% of the total area of plot) and is characterized by 20–30 m tall trees with a diameter at breast height >15 cm (Williams-Linera 2002; Flores-Palacios and García-Franco 2006) and (2) NF, which includes different land-use types resulting from human activity such as pastures, young secondary vegetation, and commercial tree plantations (Figure 1). Average precipitation during the dry season was 132 mm (monthly range: 50–300 mm) and 237 mm (80–385 mm) during the rainy season (data from an automated meteorological station EMA 30452, <https://smn.conagua.gob.mx>).

**Table 1.** Life history traits of *D. mexicanum* and *D. satanas* obtained from the literature available for the genera and related species

Life history traits	Species		References
	<i>D. mexicanum</i>	<i>D. satanas</i>	
Relocation strategy	Roller	Tunneler	Arellano et al. (2005)
Feeding habit	Copro-necrophagous	Coprophagous	Solis (1987); Verdú et al. (2006); Halffter et al. (2007)
Diel activity	Nocturnal	Nocturnal	
Body size	17.4–23.1 mm	16.0–25.5 mm	
Sexual dimorphism	Absent	Horns on head and thorax	
Nesting behavior	The pair of beetles meets in manure, builds and transports the nest ball (bisexual cooperation), which is buried at a shallow depth under the ground	Bisexual cooperation when preparing the nest ball, which is buried more deeply (>50 cm) under or near the food source	Joseph (1929); Halffter and Edmonds (1982)
Fecundity	1–3 eggs per female	2–9 eggs per female	Cambeftor and Hanski (1991); Barretto et al. (2021)
Parental investment	Present, usually performed by both sexes	Absent or reduced, performed by female	Barretto personal observation; Joseph (1929); Halffter and Matthews (1966); Halffter and Edmonds (1982)
Max. estimated population size	4,687 individuals	4,395 individuals	Barretto et al. (2018)
Max. estimated recruitment	2.6 individuals	1.2 individuals	
Max. estimated survival	6.48 individuals	5.44 individuals	
Habitat use	Forest specialist—associated with well preserved, forested areas	Generalist—associated with open and semi-open areas like secondary forests, pastures, crops, and urban areas	Escobar (2004); Rös et al. (2012); Barretto et al. (2018)

To determine between-habitat variation in temperature and humidity, we used 6 data loggers (iButton<sup>®</sup>), 3 in each habitat. In the forest, mean temperature during the dry season was 14.5 °C (range: 1–27.2 °C) and humidity was 68% (45–92%). During the rainy season, the mean temperature was 16.2 °C (4–36 °C) and humidity was 84% (76–95%). In NF, mean temperature during the dry season was 16.2 °C (4–36 °C) and humidity was 62% (46–92%). During the rainy season mean temperature was 18.6 °C (12.5–37 °C) and humidity was 79% (63–93%).

Within the 126 ha plot, we selected 36 sites separated by >50 m (16 in the forest, and 20 in NF), in order to cover as much of these 2 habitats as possible (Figure 1). These were sampled monthly over the course of 1 year (August 2015 to July 2016). At each site, a non-lethal pitfall trap was set up (Figure 1). The trap consists of a 1-L container (13.5 cm high and 9 cm in diameter) with a plastic funnel on top to prevent the beetles from escaping (Escobar and Chacón de Ulloa 2000), buried flush with the soil's surface and half-filled with soil to capture the live beetles. Each trap was baited with 50 g of human excrement placed in a 50-mL plastic cup held by a wire for capture periods of 24 h, with 1 day between sampling events to ensure the dispersal of the marked beetles (Cultid-Medina et al. 2015). Thus there were 5 days of effective sampling per month (120 h/month, in total). Human excrement is one of the most efficient attractants for capturing beetles, including necrophagous species, since their feeding preferences and reproduction are commonly associated with higher nutritional quality dung of omnivorous mammals such as humans (Bogoni and Hernández 2014).

Beetles captured in the traps were marked with a consecutive number code on the surface of the pronotum or the elytron using a Motool (Dremel Stylus 1100<sup>®</sup>) and following the protocol of Martínez-Quintero et al. (2013). For each beetle, we recorded the species, sex, the number of trajectories and distance covered, date, and the type of habitat in which the beetle had been captured and

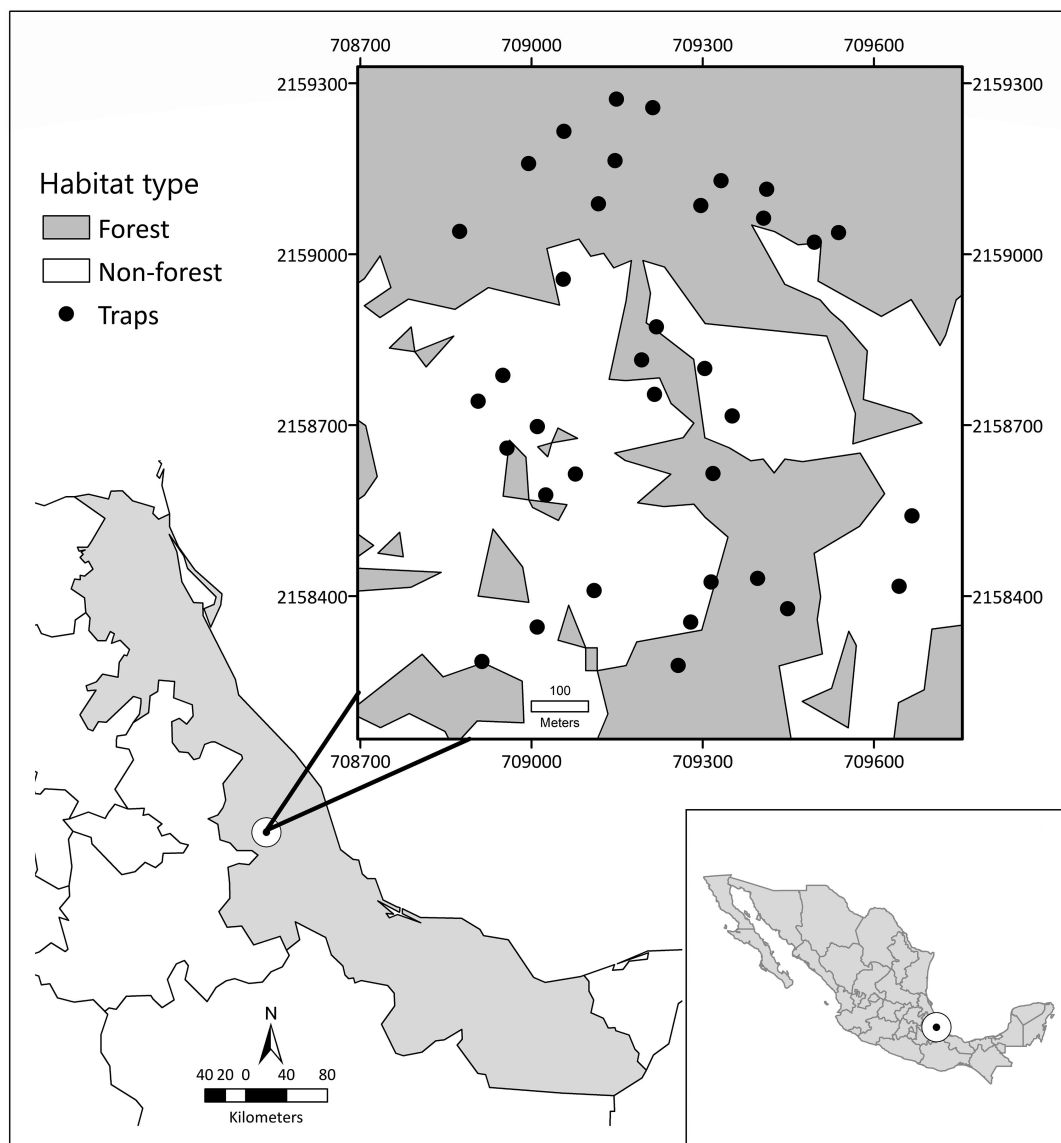
recaptured. Sex was determined based on sexual dimorphism in *D. mexicanum* (differences in the tarsal claw shape of the foreleg) and based on secondary sexual characters in *D. satanas* (presence of horns on head and thorax; Table 1). Data were only taken for beetles with a rigid, chitinized exoskeleton as these are considered mature adults or are in the process of maturation (Halffter and Edmonds 1982; Barretto et al. 2021). The capture–recapture history over time was obtained for each beetle, which in statistical terms is considered the best approach for studies at the population level (Fujiwara and Caswell 2002; Ancona et al. 2017).

## Data analysis

### Spatial and temporal variations in ASR

ASR was estimated as the number of males/(males + females) (Wilson and Hardy, 2002; Ancona et al. 2017). This estimator is symmetric around 0.5 and bounded between 0 (only females in the population) and 1 (only males in the population). Thus, on a similar scale, it is possible to obtain the same degree of bias toward both females and males. This method is recommended for estimating ASR under natural conditions since it treats individuals as independent units in the population (Ancona et al. 2017).

We calculated ASR for both species per month (over 12 months), per season (rainy and dry), and per habitat type (forest and NF). As the distributions of the observed ASR values were often non-normal, a non-parametric procedure was used to determine the asymmetric 95% confidence limits using the *percentil-t* bootstrap method (10,000 resamplings) (Efron 1982; Mooney and Duval, 1993), available in the boot library of R version 3.6.2 (R Core Team 2019). The distribution of the resampling values allowed us to evaluate the statistical significance of the ASR values. According to Cumming (2014), when the 95% confidence intervals (CIs) of independent means do not overlap, the 2-tailed *P*-value is < 0.05 and there is a statistically significant difference between the means. This method



**Figure 1** Geographic location of the study area (126 ha) showing the spatial distribution of pitfall traps (filled circle) in both habitat types in a cloud forest landscape in Veracruz, Mexico.

also allowed us to know how accurate the ASR estimate was and compare the variation in the estimated ASR within and between species. In addition, to find out if the ASR values of the 2 species depend on season and month by habitat, chi-square tests with Yates' continuity correction were run (Crawley 2007).

#### Male aggregation

We calculated male aggregation per trap using mean crowding ( $m^*$ ) as proposed by Lloyd (1967). For species that live in ephemeral resources and that are captured in baited traps,  $m^*$  is useful for measuring the degree of spatial or temporal aggregation of individuals in a manner relevant to intraspecific competition for resources (Inouye 2005; Wade et al. 2018).  $m^*$  was calculated as:

$$m^* = m + \left[ \left( \frac{\sigma}{m} \right) - 1 \right],$$

where  $m$  = mean density per trap (mean density) and  $\sigma$  = variance of mean density (variance). For both species,  $m^*$  was calculated

using the number of adult males per trap per habitat (F and NF) and for both seasons (rainy and dry). Similar to ASR, the 95% CIs for the observed  $m^*$  values were estimated using the *percentil-t* bootstrap method (resampling 10,000 times) (Efron 1982; Mooney and Duval 1993) from the boot library available in R version 3.6.2 (R Core Team 2019). To see whether the observed  $m^*$  values of the 2 species depend on sex or season by habitat type, chi-squared tests with Yates' continuity correction were performed (Crawley 2007).

#### Movement by species and by sex

To describe the movement of both species across the landscape, we used the capture–recapture data for each beetle over the 12 months of the study, taking into account the movement trajectory of each sex per species in each habitat and between habitats. Trajectory is the linear displacement of a beetle between traps and allows us to estimate the number of times a beetle moved, as well as the minimum, mean and maximum distances covered. Chi-squared tests with Yates' continuity correction were run to evaluate whether the

**Table 2.** Number of individuals by sex and total density (beetles per trap) of *D. mexicanum* and *D. satanas* captured in a cloud Forest landscape (Veracruz, Mexico) according to habitat type and season

Habitat type	Sex	Species/season							
		<i>D. mexicanum</i>				<i>D. satanas</i>			
		Rainy	Dry	Total	Density	Rainy	Dry	Total	Density
Forest (Traps = 16)	♀	279	143	422	26.3	48	16	64	3.2
	♂	225	125	350	21.8	67	24	91	4.5
	Total	504	268	772	48.2	115	40	155	9.6
NF (Traps = 20)	♀	134	38	172	10.7	197	32	229	11.4
	♂	100	25	125	7.8	221	30	251	12.5
	Total	234	63	297	14.8	418	62	480	24

number of movement trajectories of each species depended on the habitat or season (Crawley 2007).

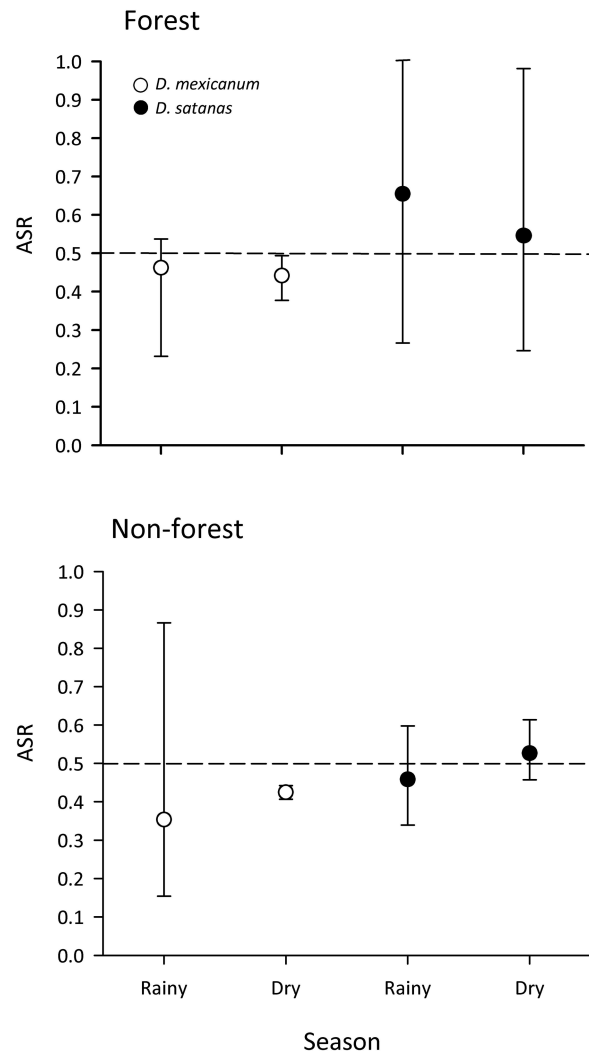
To compare the distribution of the number of movement trajectories per species and per sex in each habitat, a goodness-of-fit test for contingency tables was used with the *mosaicplot* function in the *graphics* R package (Pruim et al. 2017). This function generates the standardized residuals (SRs) of a log-linear model (SR) for the contingency table. In our case, when the proportions observed in the contingency tables differ from those expected (residual values less than  $-2$  and greater than  $2$ ), the observed number of trajectories is significantly different from that expected by chance. This method is recommended when the expected frequencies of  $\geq 1$  of the categories to be analyzed are low. Therefore, log-linear modeling fits a saturated model with zero residual deviance and then simplifies the model by removing the highest-order interaction term (Crawley 2007). For the analysis, we used R version 3.6.2 (R Core Team 2019).

## Results

A total of 1,069 adult *D. mexicanum* were marked and sexed (594 ♀ and 475 ♂). Net recapture (single recaptures) was 15% (165 beetles): 92 females (8.3%) and 73 males (6.6%). In this species, 28 beetles (12 males and 16 females) were recaptured twice. For *D. satanas*, 635 adult beetles were marked and sexed (293 ♀ and 342 ♂). Net recapture was 6% (38 beetles): 18 females (2.8%) and 20 males (3.1%) and, only 2 beetles (1 male and 1 female) were recaptured twice. The abundance and total population density (beetles/trap) of *D. mexicanum* were higher in the forest and this pattern was the opposite of that observed for *D. satanas*, in which abundance and total population density were higher in NF (Table 2). In both habitats, the species were more abundant during the rainy season (Table 2).

### Spatial and temporal variation in ASR

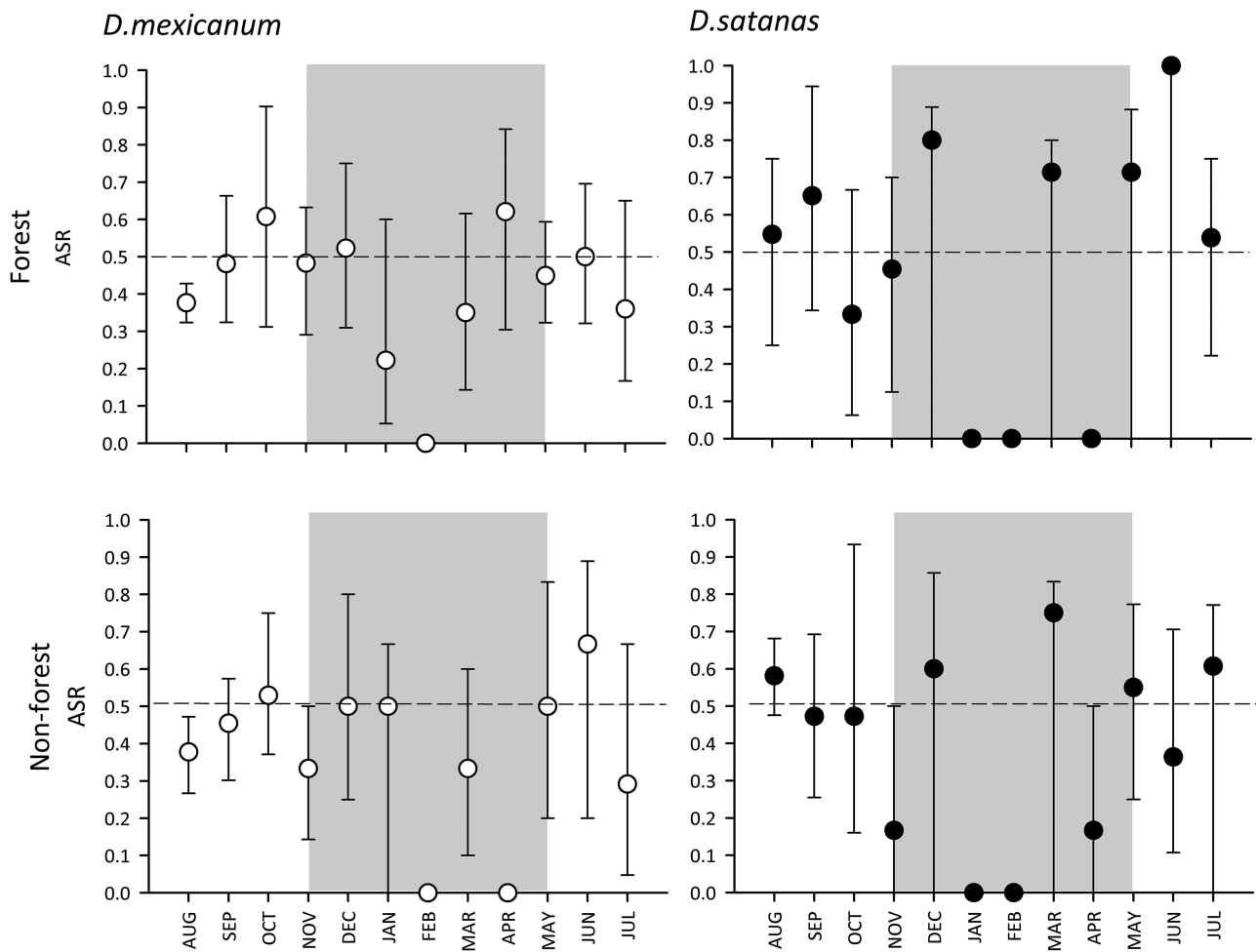
We found opposite tendencies in the observed ASR values for the 2 species studied. While the population of *D. mexicanum* tended to be female-biased, the population of *D. satanas* tended to be predominantly male, except in NF at rainy season (Figure 2). The overlap of the 95% CIs indicates that there were no significant differences in the observed ASR values (Figure 2). This is consistent with what was found when examining whether the observed ASR values were associated with habitat and season. The analysis indicated that the



**Figure 2.** Spatial and temporal variation in the ASR of *D. mexicanum* and *D. satanas* for the rainy and dry seasons in each habitat type in a cloud forest landscape, Veracruz, Mexico. The dots represent the observed ASR for each population and bars are 95% CIs estimated by bootstrap (10,000 resampling). The dashed horizontal lines at 0.5 represent the ASR in equilibrium (i.e., same number of females and males).

observed ASR values for each species in both seasons were similar and that these did not depend on habitat ( $\chi^2_{\text{dry}} = 0.01$ ,  $df = 1$ ,  $P = 0.88$ ;  $\chi^2_{\text{rainy}} = 0.54$ ,  $df = 1$ ,  $P = 0.46$ ). However, it is important to highlight that the ASR estimates are highly variable between seasons as a function of habitat type (Figure 2). For *D. mexicanum*, the range of variation in the 95% CIs of the observed ASR was greater during the rainy season, mainly in NF. In contrast to this, for *D. satanas*, the 95% CIs were more variable in the forest than in NF during both seasons; though in NF, this population tended to be female-biased during the rainy season (Figure 2).

Another interesting pattern is that despite the large monthly variation in the ASR values for both habitats, the observed ASR throughout the year depends significantly on the species ( $\chi^2_{\text{forest}} = 97.45$ ,  $df = 10$ ,  $P < 0.001$ ;  $\chi^2_{\text{NF}} = 119.70$ ,  $df = 10$ ,  $P < 0.001$ ) (Figure 3). For the *D. mexicanum* population, the observed ASR values were mainly biased toward females in both habitats, with notable variation in the 95% CIs for NF (Figure 3). The *D. satanas*



**Figure 3** Spatial and temporal ASR variation throughout the year (August 2015 to July 2016) for *D. mexicanum* and *D. satanas* in 2 habitat types (forest and NF) in a cloud forest landscape, Veracruz, Mexico. The dots represent the observed ASR for each population and bars are 95% CIs estimated by bootstrap (10,000 resampling). The dashed horizontal lines at 0.5 represent the ASR in equilibrium (i.e., same number of females and males). The dry season is in gray.

population was biased toward males in forest and to female in NF, and in the forest the 95% CIs values were more variable (Figure 3).

#### Male aggregation

Spatio-temporal male aggregation ( $m^*$ ) was also variable between species. As shown by the nonoverlap of the 95% CIs, *D. mexicanum* and *D. satanas* were significantly more aggregated during the rainy season in both habitats, except *D. satanas* in forest, for which the aggregation of males was similar between seasons (Figure 4). This was corroborated when examining whether male aggregation in each season was associated with habitat type. In the rainy season, male aggregation in both species was significantly dependent on habitat ( $\chi^2 = 5.24$ ,  $df = 1$ ,  $P = 0.02$ ), but in the dry season, male aggregation was independent of habitat type ( $\chi^2 = 1.33$ ,  $df = 1$ ,  $P = 0.24$ ).

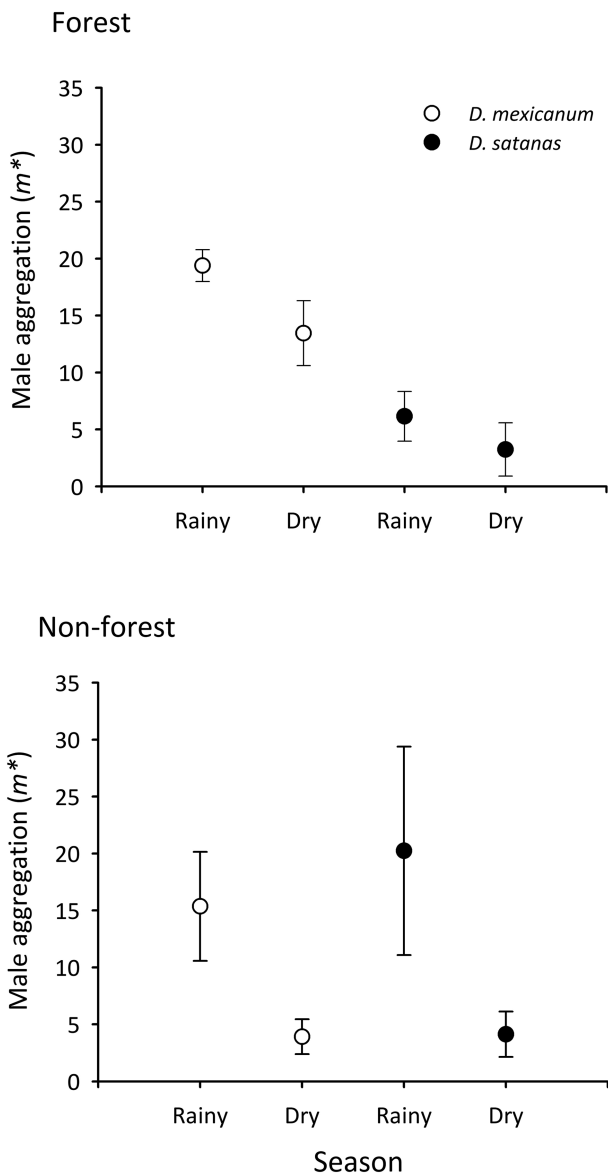
#### Movement by species and by sex

Throughout the year, we recorded a total of 193 trajectories for *D. mexicanum* and 40 for *D. satanas*. For the sexes of both species, the number of movement trajectories depends on habitat (females:  $\chi^2 = 27.85$ ,  $df = 2$ ,  $P < 0.001$ ; males:  $\chi^2 = 28.78$ ,  $df = 2$ ,  $P < 0.001$ ). Fifty-eight percent of the movement trajectories of *D. mexicanum* were limited to the forest, 7% were limited to NF, and 35% took

place between habitats (Figure 5). In this species, both sexes moved fewer times in NF than expected by chance. For *D. satanas*, 23% of the trajectories were limited to the forest, 50% limited to NF, and 27% took place between habitats (Figure 5). Contrary to what was found for *D. mexicanum*, *D. satanas* moved more than expected by chance in NF (Figure 5). On comparing the number of movement trajectories per season, no statistical differences were found ( $\chi^2 = 0.03$ ,  $df = 1$ ,  $P = 0.864$ ). The mean linear distance ( $\pm$ SE) covered by male *D. mexicanum* was  $282 \pm 33$  m and that of females was  $225 \pm 24$  m. In this species, the maximum distance covered was 1,000 m in 48 h by 2 beetles. Male *D. satanas* covered a mean linear distance of  $278 \pm 49$  m and females,  $285 \pm 99$  m. The maximum linear distance recorded was 1,660 m over 33 days by a male.

#### Discussion

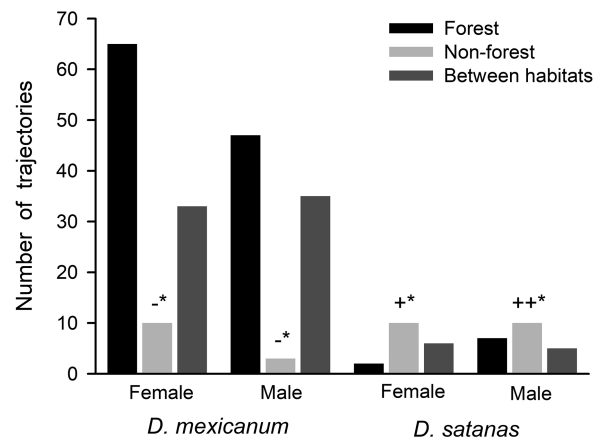
Through capture–recapture over the course of a year for 2 species of dung beetle that coexist in a cloud forest landscape, we compared the spatiotemporal variation in their ASR, male aggregation ( $m^*$ ) and movement patterns; all of which are ecological parameters that have received little study in natural populations of insects. Four findings deserve special attention. (1) There were, as we predicted, opposite patterns in the ASR as a function of the 2 species' life history



**Figure 4.** Spatial and temporal mean crowding ( $m^*$ ) calculated for males of *D. mexicanum* and *D. satanas* populations for 2 seasons (dry and rainy) in 2 habitat types (forest and NF) in a cloud forest landscape, Veracruz, Mexico. Dots represent the mean calculated value of  $m^*$  and bars the estimated 95% CIs by bootstrap (10,000 resampling).

traits, and observed ASR values were not related to habitat type or season. Despite this, the 95% CIs of ASR accuracy varied with habitat and season. (2) Monthly ASR was variable and depended on the species. (3) As we expected, the males of both species were more aggregated in the rainy season, when the emergence of new individuals and reproduction occurred. However, the magnitude of male aggregation varied between species according to habitat type. (4) The movement patterns of these species across the landscape depended on their habitat preferences and were similar between seasons.

It is known that repeated sampling of marked individuals produces reliable estimates of many population parameters, and Ancona et al. (2017) point out that capture–recapture is one of the most appropriate methods for estimating sex ratio under natural conditions. Compared with other studies on dung beetles at the population level



**Figure 5.** Total number of movement trajectories for each sex of *D. mexicanum* and *D. satanas* recorded in both habitats and between habitats (forest and NF) in a cloud forest landscape, Veracruz, Mexico. In detail, the values of SRs, which were significantly lower (–\* SR = –4–2) and greater (+\* SR = 2:4, ++\* SR < 4) than expected by chance.

(Escobar and Chacón de Ulloa 2000; Escobar 2003; Arellano et al. 2008; Silva and Hernández 2015; Gray et al. 2021), we recorded a high recapture rate (6–15%) and this allowed us to reliably describe the variation in ASR as well as the aggregation of males and movement patterns for both species. Although biases in catchability and the probability of detection due to physiological state and behavioral differences between the sexes has been found in several taxa (Dénes et al. 2015), in dung beetles, there is no sufficient evidence that baited traps attract one sex more than the another. However, recently Salomão et al. (2021), on comparing 2 species of *Dichotomius*, showed that in *Dichotomius iannuzzie* food attraction was related to sex and age, while in *Dichotomius gillettei* attraction did not depend on sex or age. This is a topic that has received little attention in dung beetles and that should be the subject of future studies on population ecology in this group of insects.

Examining changes in ASR is critical to be able to predict the intensity of competition for mates and resources and is therefore essential for understanding the evolutionary trajectories of wild populations (Hardy 2002). Some studies on dung beetles suggest that the female bias and aggregation of males can reduce the level of direct competition among males (Moczek 2003; Pomfret and Knell 2008). According to Sapir et al. (2008), when male–male competition is minimized, natural selection favors an offspring sex ratio that is highly female-biased. In *D. mexicanum*, a hornless species, female dominance in the population, and the greater aggregation of males appear to be compensatory responses to the cost of producing few oocytes (between 1 and 3, see Table 1). This should result in an increased probability of mating and a greater rate of fertilization per female (Jennions and Fromhage 2017), favoring parental investment (Halffter and Edmonds 1982), as seems to occur in this species. This suggests that, as observed in species with scramble competition polygyny (Baena and Macías-Ordóñez 2015), there could be less interference competition among males (i.e., confrontations or fights) in *D. mexicanum* as a result of the greater abundance of females, or owing to their being widely dispersed, making monopolization by the males difficult.

In contrast, for *D. satanas*, a species with horns, the ASR was male-biased and the spatial and temporal aggregation of males was lower, except in NF at rainy season. In reproductive terms, the bias toward males in *D. satanas* indicates greater intrasexual competition

given that females were proportionally less abundant, they produce a greater number of oocytes (2–9; Barretto et al. 2021), and both sexes apparently make a smaller parental investment (Halfpeter and Matthews 1966; Halfpeter and Edmonds 1982). In addition, a lower number of female dung beetles have also been associated with an increase in intra and interspecific competition that limits the availability of the resources needed for feeding and reproduction. Consequently, females may not be able to achieve maximum fecundity (Moczek 2003). However, during the rainy season when reproduction occurs, the ASR female-biased at NF seems to be a strategy to increase the fecundity. These adjustments have consequences for population size and density, as found in *D. satanas* (Barretto et al. 2018).

Our results show that even though each species exhibited biases toward a specific sex, habitat and season did not influence ASR trends. However, the range of accuracy for estimated ASR and the influence of seasonality on male aggregation are noteworthy. Several studies have indicated that a higher degree of aggregation and sex ratio biased toward the more common sex are usually found in habitats and during the seasons that offer the better conditions for the survival and reproduction of individuals (McKellar et al. 2009; Reichard et al. 2014). For dung beetles, the reproductive season coincides with an increase in temperature and precipitation during the rainy season (Halfpeter and Edmonds 1982; Barretto et al. 2021). The highest aggregation of males was observed for both species during this season, as was the greatest variation in ASR. For example, in NF, the ASR of *D. mexicanum* was highly variable and for *D. satanas* it tended to be biased toward females in the rainy season. Temporal adjustments in population parameters seem to be a response that guarantees the reproductive success of the rarer sex by increasing the rate of mating during the reproductive season (Pettersson et al. 2004; Kokko and Rankin 2006). This is also supported by the large monthly variation in ASR values of both species probably as a function of reproductive phenology, which could be related to changes in the availability of resources throughout the year. However, the impact of annual variation in food quantity and quality on reproductive phenology and demographic parameters of dung beetle populations has received little attention.

Changes in the demographic parameters associated with environmental conditions have also been found to be related to dispersal and the differential mortality of the sexes, with the prediction that the ASR in better quality habitats will be biased toward the sex with a lower capacity for movement and a lower probability of death (Julliard 2000). In our study, movement was different between species and was related to differential habitat use and their resource exploration ability, as expected for species with opposite sets of ecological traits (Cultid-Medina et al. 2015; Silva and Hernández 2015; Gray et al. 2021). In this sense, our results—supported by the greater rate of recapture, number of trajectories, and distance covered within the forest—show that female *D. mexicanum* are the more common sex and according to Barretto et al. (2018), the one with a lower probability of death. In contrast, the low recapture rate for *D. satanas* indicates this species spent less time in the forest and was more mobile in NF areas. The results also show that the rate of recapture and the distance covered by *D. satanas* are similar for both sexes, suggesting that both males and females have the same capacity for movement and/or a similar probability of death. This validates the proposal of Barretto et al. (2018), who state that the differences in the demographic parameters between species are due to differences in their life history traits and movement patterns, which affect the balance between the entry and exit (i.e., dispersal

and migration) processes of individuals in the population, directly influencing ASR and aggregation (Julliard 2000).

Our results show that, rather than a trend toward balanced sex ratios (1:1), in both dung beetle populations, there were biases in ASR and differences in the aggregation of the males related to the life history traits of the species, with consequences for population dynamics. Even though ASR did not depend on habitat or seasonality, the range of variation in ASR was less in habitats where both sexes of each species dispersed more, and this variation was greater where they were less frequent. The high degree of variation in ASR and greater male aggregation in the rainy season when emergence and reproduction coincide with the highest temperature and humidity, could be a cue for adaptive plasticity regarding how each sex adjusts its survival strategies relative to ecological and reproductive resource availability. Furthermore, it is remarkable that the observed changes in ASR were more short-term than long-term, indicating that the adjustment in ASR is dynamic over time in response to environmental variation. In this sense, our results elucidate aspects of how species use and explore different habitats and how variation in demographic parameters have implications for the ecology and evolution of the species studied, relative to the dynamics of the entry and exit of individuals through differential species-specific adjustments that can influence the dispersion and growth of populations. We argue that comparative population studies on species with different life history traits are necessary to examine variation in demographic parameters to understand ecological and evolutionary implications in the face of spatial and climatic environmental variation.

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## Authors' Contributions

J.B., M.L.B., and F.E. conceived the study. J.B. conducted the field work and collected the data. J.B., M.L.B., and I.H.D. performed the statistical analyses. All authors contributed to the writing of the manuscript.

## Conflict of Interest

The authors declare no conflict of interest.

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