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Point-count methods to monitor butterfly populations when traditional methods fail: a case study with Miami blue butterfly

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Abstract Established butterfly monitoring methods are designed for open habitats such as grasslands. Not all rare species occupy habitats that are easy to see across and navigate, in which cases a new approach to monitoring is necessary. We present a novel use of point transect distance sampling to monitor the Miami blue, a highly endangered butterfly that occupies dense shrub habitat. To monitor Miami blue density, we developed surveys consisting of butterfly counts in semi-circular plots. We examined the rate at which an observer detects new butterflies to determine the survey duration that meets the key assumption that butterflies are detected at their initial location. As a related secondary goal, we identified the determinants of adult flight phenology to target monitoring efforts during periods of high adult abundance. We observed peak Miami blue densities in April and July/August 2012, and July/August 2013. We estimated density using detections from a 10-sec survey, our most defensible and conservative estimate. Peak daily density estimates ranged from 592 to 680 butterflies per hectare. Adult density was related to precipitation patterns, with high densities occurring 4–6 weeks after particularly wet 4-week intervals. For butterfly species that exist in high enough densities, we recommend using point transect distance sampling in habitats where traditional methods are impossible to implement.

Keywords Conservation · Distance sampling · Subtropics · Endangered species

Introduction

Rigorous monitoring and abundance estimates for rare insects are essential to conserve and recover their populations. However, defined monitoring protocols and rigorous abundance estimates are lacking for many endangered species, including a number of rare butterflies. The most rigorous approaches to population estimation: mark-recapture and transect counts (Brown and Boyce 1998; Haddad et al. 2008), are not always possible to implement. We implement a new approach for butterfly population estimation, based on distance sampling from point transects, defined as transects of zero length (Buckland et al. 2001), that can be applied when assumptions of traditional methods fail.

The most common methods used for monitoring butterfly populations are mark-recapture and transect counts. Mark-recapture methods are the most rigorous because they allow for estimation of daily and total population sizes, recruitment, survival, and detection probabilities (Haddad et al. 2008). However, these methods are resource intensive and have the potential to harm fragile butterflies in the marking process (Murphy 1987). As an alternative, transect counts are commonly used to monitor butterfly populations because they are non-invasive, have a long history in population and community monitoring, and allow for estimation of detection probabilities by way of

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distance sampling (Pollard 1977; Thomas 1983; Brown and Boyce 1998; Isaac et al. 2011). Distance sampling from line transects is increasingly used to estimate butterfly density and abundance of rare species (Brown and Boyce 1998; Hamm 2013) as well as butterfly metapopulation structure, habitat associations, and response to habitat restoration treatments (Boughton 2000; Powell et al. 2006; Pocewicz et al. 2009; Moranz et al. 2014). Transect counts are easily deployed in open habitats such as grasslands, but can be problematic in dense vegetation where it is difficult to simultaneously count butterflies and navigate. Adding distance sampling to transect counts is also complicated by dense vegetation because of the need for transect locations to be random (Buckland et al. 2001). Another method used to incorporate detection probability into abundance estimates is occupancy modeling (Bried and Pellet 2012; van Strien et al. 2013). One of the fundamental assumptions of this method, however, is that a population is closed between survey visits (Mackenzie et al. 2006), an assumption that may be hard to satisfy for butterfly species that are logistically difficult to access, have overlapping broods, and are surveyed sporadically. We overcome the limitations imposed by other, more traditional butterfly monitoring approaches and develop methods for point transects for insects, adapting methods originally developed for estimating bird abundance (Rosenstock et al. 2002; Buckland 2006).

We develop our new approach using a case study of an extremely rare and threatened butterfly, Miami blue (*Cyclargus thomasi bethunebakeri*). Miami blue is a US Federally Endangered species (USFWS 2012) for which rigorous monitoring protocols and abundance estimates have yet to be developed. Traditional monitoring approaches are nearly useless for these butterflies; Miami blues are small (19 mm wingspan) and occupy remote, uninhabited islands at the western end of the Florida Keys (Fig. 1). Access to these islands is often impossible because they can only be approached by small boats dependent on favorable weather and tides. Additionally, Miami blue habitat is characterized by dense, shrub-dominated, coastal thickets. These thickets are bordered on the inland side by impenetrable mangroves and on the seaward side by coastal prairie composed of vines, sand-spurs, waist high sea oats, and old hurricane debris. All three habitat types are particularly difficult to traverse. Although trails could be created, these would harm butterfly habitat, and would provide access points for illegal visitors to restricted wilderness islands. These size and habitat restrictions make Miami blue an ideal candidate for testing new abundance estimation methods.

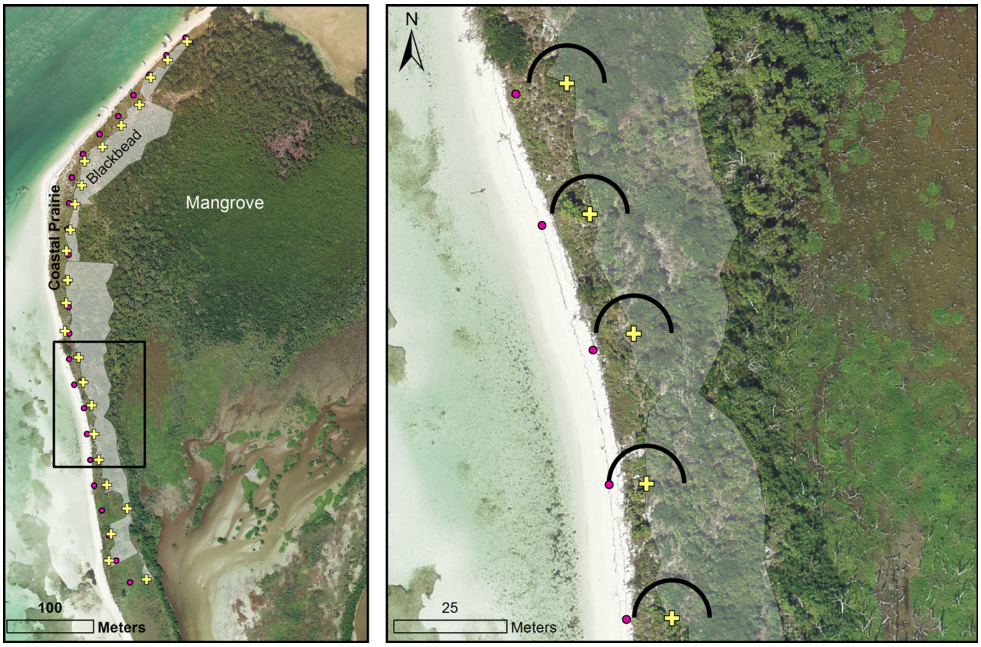
A second goal of our study is to make the monitoring protocol we develop more efficient by pinpointing periods of high adult flight activity within years. Nearly all established butterfly monitoring protocols are developed for species in temperate regions where butterflies exhibit predictable phenology patterns. Generally, temperate butterflies fly as adults in spring and summer, and diapause during winter months when cold temperatures limit insect development. This makes it relatively easy to target monitoring efforts during periods of high adult abundance. In the sub-tropics and tropics, however, temperatures vary little and remain warm throughout the year. Instead, it is precipitation that varies annually with distinct dry and rainy seasons. Tropical butterfly responses to seasonality are varied, with some species flying at high abundances at the start of the rainy season, others at the end of the rainy season, and still others flying during the dry season (Wolda 1988). This presents a considerable challenge for monitoring sub-tropical butterflies, especially if the phenology of a species is poorly understood and occupied habitat is difficult to access. The limited data that exist for the extant populations of Miami blue butterflies suggest a high amount of intra- and inter-annual variation in flight phenology (Daniels 2010; Cannon et al. 2010; Henry et al. 2012). We investigate the relationship between rainfall and Miami blue abundance, developing a predictive model of adult flight to streamline future monitoring efforts.

Methods

Study species and sites

Miami blues were once abundant in south Florida, especially in the area in and around Miami and throughout the Florida Keys (Saarinen and Daniels 2012). Miami blue habitat was destroyed as Miami, in the first half the twentieth century, and then the Keys, in the second half, developed. By 1992, there were so few Miami blues that the butterfly was presumed extinct when Hurricane Andrew destroyed what was then the last known population on Key Biscayne in the Florida Keys. The butterfly was re-discovered at Bahia Honda State Park (hereafter, Bahia Honda) in 1999 (Calhoun et al. 2002), and in the Key West National Wildlife Refuge (hereafter, Key West Refuge) in the winter of 2006 (Cannon et al. 2010). Because of its accessibility, the Bahia Honda population was the subject of the majority of Miami blue research efforts until the population went extinct in 2010 (Saarinen and Daniels 2006; Saarinen et al. 2009; Trager and Daniels 2011). Very little is known about the butterflies in Key West Refuge because the islands occupied by Miami blues are 25–40 km west of Key West and only accessible by small boat. Winter winds and summer thunderstorms greatly limit the number of days that it is possible to access the islands to study the butterflies. Existing, limited, data reveal notable

Fig. 1 Example Miami blue butterfly habitat and survey locations. Coastal prairie, blackbead and mangrove habitats are labeled. Dots represent locations mapped every 30 m along the beach, crosses represent actual survey locations, and semi-circles have a radius of 8 m, the farthest detection distance in our data set



differences between butterflies at the Key West Refuge and Bahia Honda. At Bahia Honda, Miami blues used nickerbean (*Caesalpina bonduc*) as the larval host, and peak adult counts were observed during the summer (Emmel and Daniels 2008). In contrast, populations in the Key West Refuge use blackbead (*Pithecellobium keyense*) as their host plant, and preliminary data (pre 2012) suggested that adults were most abundant during the winter/spring (Henry et al. 2012).

In the Key West Refuge, Miami blues occupy seven beaches on five islands including Boca Grande Key and the Marquesas Keys (Cannon et al. 2010). These beaches range in length from 250 m to 3 km (Table 1) and contain most of the undeveloped, upland coastal berm habitat in the Florida Keys. The coastal berm is dominated by blackbead, the butterfly's host, which grows in dense thickets (FNAI 2010). This impenetrable, linear strip of blackbead ranges in width from one shrub to 60 m, and individual shrubs can grow up to 4 m tall. Female butterflies lay eggs singly on newly emerging leaves and flower buds of blackbead. The distribution of the butterflies is closely tied to the distribution of blackbead on the islands.

Counts of adult butterflies

We conducted Miami blue surveys as often as weather conditions allowed boat access to the Key West refuge. Our goal was to survey each occupied beach a minimum of once per month from March 2012 through December 2013. On each visit, we performed point transects along the length of each beach bordered by blackbead-dominated coastal berm. On four beaches, this encompasses the entire length of the blackbead. On the other three beaches the blackbead extends behind the mangroves, beyond the reach of the beach. To maximize our time spent surveying on these islands, we limited our survey to the blackbead accessible from the beach.

We defined a survey "point" as the centroid of the location where we conducted our butterfly counts. To select our survey points, we used ArcGIS 10.0 (ESRI, Redlands CA 2010) to determine a line parallel to the beach extending the length of blackbead habitat. We then marked a location every 30 m along that line. For each visit, we chose a new, random starting point along the line and generated new point locations. Upon arriving at a location

Table 1 Name, length, number of survey points, and estimated habitat area for each occupied beach

	Boca grande	East	Snook	Short	Third	Main	Long	Total
Beach length (m)	1100	221	529	503	638	1023	2800	6814
Number of survey points	24	8	7	9	15	22	100	185
Habitat area (ha)	1.8	2.9	0.35	1.7	3.6	2.8	5[a]	18.15

[a] Habitat area was physically mapped on all beaches except Long where it was estimated using known blackbead locations and aerial imagery

along the beach, we walked inland until we intersected the blackbead edge. This point at the edge of the blackbead was our survey point. On all beaches except the longest (Long Beach), we surveyed all points on each visit, identifying the location of the point with a handheld GPS. Because all points on Long Beach could not be surveyed in 1 day, we randomly selected 30 of the 100 points each visit and surveyed as many of those as time/tides/weather allowed.

Most of the butterflies we counted were first detected in flight, therefore, we restricted our survey area to a semi-circle. We oriented the flat edge of the semi-circle perpendicular to the shrub edge to incorporate both dune grass and blackbead habitat. Restricting our survey to a semi-circle allowed the observer to keep track of individual butterflies, thus reducing the likelihood of double counting individuals during the survey. At each survey point, we recorded all Miami blues, Cassius blues (*Leptotes cassius*) and unknown blues (butterflies we could not confidently identify as Miami or Cassius blue) that we detected within the semi-circle during a 1-min period. Cassius blues and Miami blues look so similar that the common species, Cassius blue, is listed as Threatened under the US Endangered Species Act within the range of Miami blue (USFWS 2012). By recording Cassius blue detections we were able to include Cassius density as a covariate that might influence our ability to accurately detect Miami blue butterflies.

Distance sampling involves three key assumptions (Thomas et al. 2010) that we were able to address through our methods. First, butterflies at the point must be detected with certainty. To reduce the risk of flushing butterflies from the point, we approached survey points by walking toward the flat edge of the semi-circle, avoiding the area to be surveyed. We also started our survey the moment we arrived at the survey point and recorded any butterflies flushed from the point upon approach as detected at the start of the survey period.

A second assumption of distance sampling is that butterflies are detected at their initial location (Thomas et al. 2010). For point transects it is important to select an appropriate time interval to assure that butterflies at the point and close to the observer are detected without allowing much movement of butterflies towards the observer. The longer the duration of a point count, the more likely that butterflies move from their initial location towards the observer where they are more likely to be detected, resulting in overestimation of density (Buckland 2006). Because point transects have not been previously used to monitor butterfly populations, we recorded the time that each butterfly was detected and examined the relationship between survey duration and density.

A third assumption of distance sampling is that distance measurements are exact (Thomas et al. 2010). When we detected a butterfly, we visually estimated distance from observer in 1-m intervals. This was the smallest distance interval we were confident we could accurately estimate visually. We did not restrict the radius within which we included butterfly observations; with fixed radius counts, observers often overestimate the number of the target species present by pulling detections into the specified radius (Simons et al. 2007).

Detection probability can be influenced by a number of covariates related to climatic and habitat conditions (Dover et al. 1997; Wikström et al. 2008). In our case, we expected that blackbead cover, total shrub cover, and wind speed were all likely to influence our ability to detect Miami blue butterflies. Therefore, at each point, we estimated blackbead and shrub cover using modified Daubenmire cover classes (Daubenmire 1959), and estimated wind speed using the Beaufort scale. Temperatures below 17 °C and hours outside of peak sun can also result in reduced detectability in temperate zones (Wikström et al. 2008). In our case, all surveys occurred at temperatures above 23 °C and within a relatively narrow time window (10:00–15:30) so we did not expect these factors to dramatically influence detectability and therefore did not include them as covariates in our models.

Estimating density

To determine the survey duration that best represents the butterflies initially present at a survey point, we investigated the rate at which detections accumulated during 1-min surveys. Over time, we expect the number of new butterflies observed in a given time period to stabilize, representing the constant addition of butterflies moving into the observer's field of view. To estimate this time, we broke our 1-min survey into 5-sec intervals and determined how many new butterflies were detected in each interval. We then identified the time at which the number of new butterflies per 5-sec interval leveled off, and set our survey duration at the next time lower than that time interval.

To obtain estimates of detection probabilities (p–the probability that a butterfly in the survey area is detected) from point transects, we fit a function to our detection data that describes the observed decline in the number of detections with distance from observer. This detection function can be used to estimate $p(0)$ and the distance at which we miss as many butterflies as we detect (the effective detection radius) (Buckland et al. 2001). Using these parameters, we can estimate the density of Miami blues. Buckland et al. (2001) recommend a sample size of at least 40–60 detections to accurately fit a detection function to the data. Because we never detect 40–60 butterflies on any individual survey, we pooled data from all points surveyed across all 7 beaches during time periods when we observed peak densities. We binned our data at the following cut-points– 2.5, 3.5, 4.5, 5.5, and 6.5 m—and truncated the data at 6.5 m. By having a large interval close to the observer (0–2.5 m) we can account for movement of butterflies prior to detection at close distances where movement is more likely to affect estimation of detection probability (Buckland et al. 2001). We tested the following key function and adjustment combinations for our detection function: half-normal model with cosine adjustment, half-normal model with hermite polynomial adjustment, and hazard rate model with simple polynomial adjustment (Thomas et al. 2010). We used Akaike's information criterion (AIC) values, a Chi square goodness-of-fit test, and coefficients of variation to select the best model (Thomas et al. 2010).

By pooling data across multiple beaches, we assume that the detection function does not vary between beaches. This is biologically reasonable because the habitat on each beach is similar and survey methods are constant. Still, we assessed whether this was a valid assumption by fitting data from individual beaches to the same model we used for the pooled data set. We then compared the AIC value from the pooled model to the sum of the seven individual AIC values (Buckland et al. 2001).

Once we established our final detection function, we included the following covariates in our analysis: blackbead cover, total shrub cover, wind speed, and Cassius blue butterfly detections. We added each covariate to our detection function individually and compared AIC values to determine the best model. We then applied this model to the discreet time periods when we observed the highest Miami blue densities to compare densities across peak periods.

We fit all models in Program DISTANCE 6.0 (Thomas et al. 2010). DISTANCE calculates density by dividing the number of detections by the total area surveyed (scaled by the detection function). For a point survey with a full circle (such as for birds), the area surveyed for point transects is $k\pi w^2$ where $k =$ number of survey points and w is the effective detection radius. Because we conducted surveys over semi-circles, we multiplied the density estimated in DISTANCE by two. Confidence intervals were estimated analytically and are the sum of the variation in encounter rate (n/k) and variation in detection probability.

Estimating area of suitable habitat

Daily estimates of Miami blue population size require estimates of population density, as well as estimates of the area of suitable Miami blue habitat. Because we know very little about the habitat requirements for Miami blues, we broadly defined potentially suitable habitat as the area that is occupied by its hostplant, blackbead, within each key in the currently known range. On all beaches except for Long beach, blackbead is mostly contiguous and contained. On these beaches, we physically mapped blackbead with a handheld GPS. On Long beach, blackbead is less contained and it is impossible to physically map its entirety. Instead, we used aerial imagery to delineate the portion of the coastal berm that is mostly dominated by blackbead based on known blackbead locations.

Predicting adult flight

Early Miami blue surveys in the Key West Refuge conducted by United States Fish and Wildlife Service, Florida Fish and Wildlife Conservation Commission, and University of Florida personnel revealed high variability in the timing of adult flight periods. For example, on February 3, 2007 Cannon et al. (2010) counted 431 Miami blue butterflies on Boca Grande; the following February, Daniels (2010) counted only three butterflies on the same beach. We observed a similar phenomenon in 2012 and 2013 with peak abundances occurring at different times each year.

Two environmental factors that varied substantially between 2012 and 2013 were precipitation amount and seasonality. The spring months of 2012 were much wetter than in 2013. By the end of April 2012, Key West had received 313 mm of rain, whereas by the end of April 2013 Key West had received only 160 mm. To investigate the relationship between precipitation and Miami blue abundance, we acquired daily precipitation data from the Key West Airport weather station—the weather station closest to Miami blue habitat—through the National Climate Data Center (www.ncdc.noaa.gov). We examined the correlation between observed Miami blue densities and cumulative precipitation. We varied the interval over which precipitation was accumulated by (1) summing precipitation in week-long intervals up to 10 weeks prior to a survey and (2) moving the starting date of the summed interval back in 1 week time-steps prior to the survey date, up to 10 weeks. Each combination of these two strategies resulted in a sum of precipitation over a given length of time starting a given number of days prior to a survey. We correlated that sum of precipitation to the number of Miami blues observed per point on each survey date.

Results

We made a total of 79 trips to the Key West National Wildlife Refuge between March 2012 and November 2013, during which we surveyed a total of 1991 points. We surveyed at least one beach each month, all beaches were surveyed at least 15 times—the most accessible was surveyed 26 times. We observed three distinct periods of peak abundance: late March—early April 2012, late July—early August 2012, and late July—early August 2013 (Fig. 2).

Estimating density

Longer observation periods resulted in higher densities. Thirty percent of Miami blue butterflies observed during 1-min point counts were detected during the first 5 s of the survey. At the end of 10 s, we had detected 40 % of butterflies. After that, butterfly detections accumulated at a fairly constant rate of 6–8 % of the total detections every 5 sec (Fig. 3). Based on this analysis, we chose 10 s as our survey duration as it resulted in the most conservative, minimum estimates of population size.

Using pooled data from 10-sec surveys, the detection function with the lowest AIC_C was a half-normal model with a cosine adjustment. When we fit the data from individual beaches to this model, the combined AIC_C was greater than the AIC_C of the pooled model ($\Delta\text{AIC}_\text{C} = 7.4$). This favors our approach of pooling data from multiple beaches to estimate density. Adding covariates to the model did not have strong effects on the AIC_C values or densities; all models, except for the model that included Cassius blue density, had $\Delta\text{AIC}_\text{C}$ values <3 (Table 2) and estimated butterfly densities only varied by one Miami blue per hectare. The high $\Delta\text{AIC}_\text{C}$ of the Cassius blue model indicated that Miami and Cassius blue butterfly densities did not co-vary. Additionally there was no relationship between daily Miami blue and Cassius blue counts per point ($r^2 = 0.020$) indicating that the presence of the two butterfly species did not affect the observer's ability to correctly identify each species.

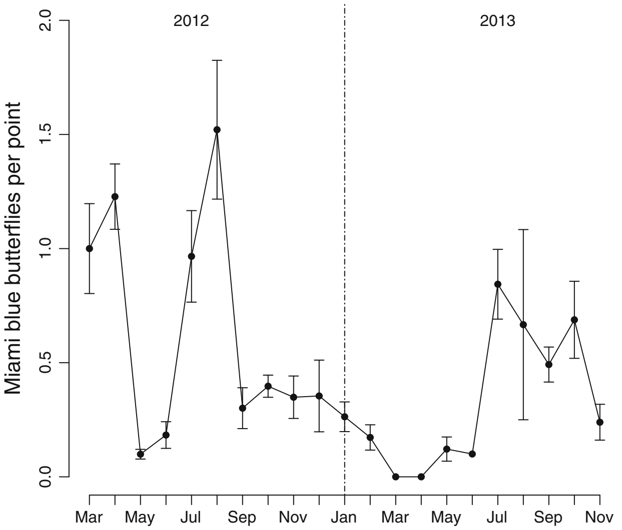


Fig. 2 Phenology of Miami blue butterfly adults in 2012 and 2013. Values are mean butterfly count per point ± SE

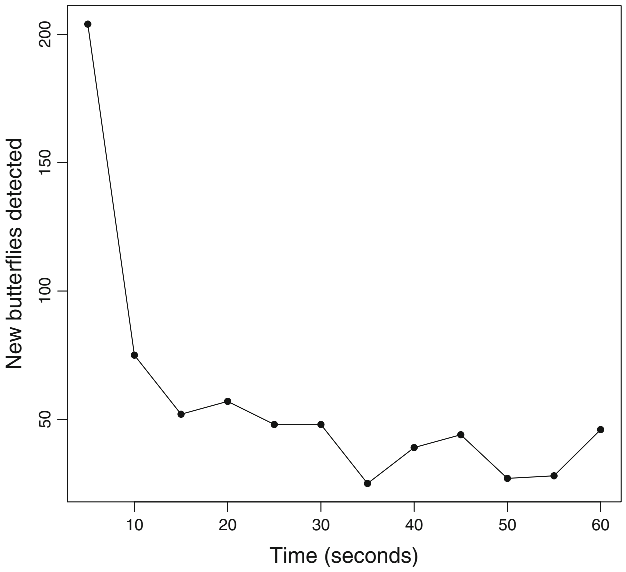


Fig. 3 The number of new butterflies detected in each 5-sec interval during a 1-min survey. The total number of detections is equal to all butterflies detected during surveys in 2012 and 2013

Table 2 Delta AIC values for detection functions with covariates

Model	# par	X^2 *p* value	Δ AIC_c	AIC_w
Null	2	0.454	1.66	0.201
Blackbead	3	0.210	2.55	0.129
Total shrub	3	0.212	0.00	0.461
Wind speed	3	0.210	1.60	0.207
Cassius density	2	0.001	13.12	0.001

par, number of parameters in the model; X^2 p-value, result of X^2 Goodness-of-fit test

Density estimates from all three peak periods were similar. In spring and summer 2012, we estimated the density to be 592 (95 % CI 500–702) and 680 (95 % CI 574–806) Miami blue butterflies/hectare, respectively. In early 2013, we observed very low Miami blue activity, until we again detected high densities in the mid-late summer. For this period, we estimated density to be 554 (95 % CI 468–656) butterflies/hectare. The coefficient of variation for these peak estimates is 0.086. DISTANCE estimated detection probability to be 0.12 (95 % CI 0.099–0.14) and the effective detection radius to be 2.22 m (95 % CI 2.04–2.42).

Estimating area of suitable habitat

We estimated a total of 18 hectares of upland coastal berm in the Key West National Wildlife Refuge (Table 1).

Predicting adult flight

The best predictor of adult density (Miami blues/point) was the sum of precipitation over the 49-day period ending 28 days prior to the survey date ($r^2 = 0.42$, $p < 0.001$; Figs. 4, 5). Once cumulative precipitation over a 49-day period exceeds 130 mm, we expect to observe Miami blue densities greater than one butterfly per point 28 days later.

Discussion

Point transect distance sampling provides an efficient method to obtain reliable and statistically rigorous estimates of butterfly densities. Whereas point counts have been used previously to estimate butterfly species richness in tropical forests (Sparrow et al. 1994), we demonstrated that by adding distance sampling we are able to effectively estimate density. This method is especially useful in butterfly habitats like those dominated by shrubs, wetland, or dense forest, where navigating transects is difficult. For example, the US Endangered St. Francis' satyr (*Neonympha mitchellii francisci*) occupies early successional wetlands dominated by sedges and small hardwood saplings (Kuefler et al. 2008). Traversing this habitat involves wading in sometimes knee-deep muck, and tramples sensitive wetland vegetation, including the butterfly's host. Using point transect distance sampling to estimate density in these habitats would allow for the estimation of detection probability and subsequent abundance with minimal damage to the habitat. These methods are not only useful for butterflies, but could be applied to insects more broadly, especially for other species that can be injured during marking and for which transect counts are logistically difficult, such as damselflies (Cordero-Rivera et al. 2002). Mark-recapture remains an important method for estimating demographic parameters and total population size. However, point transect distance sampling is a good alternative for long-term monitoring because it is less invasive, relatively cheap and easy to implement.

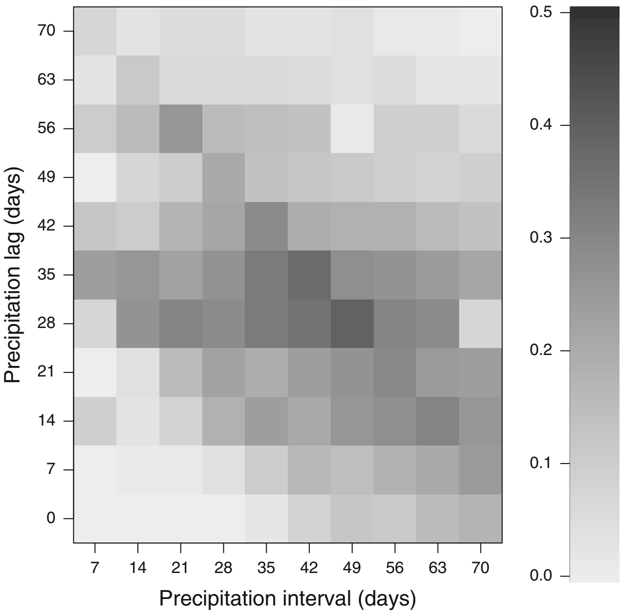


Fig. 4 Matrix of r^2 values for correlations between precipitation and Miami blue butterfly density. Each correlation uses a different number of days over which precipitation is summed (interval) ending a different number of days prior to the survey date (lag). *Darker squares* equal higher r^2 value

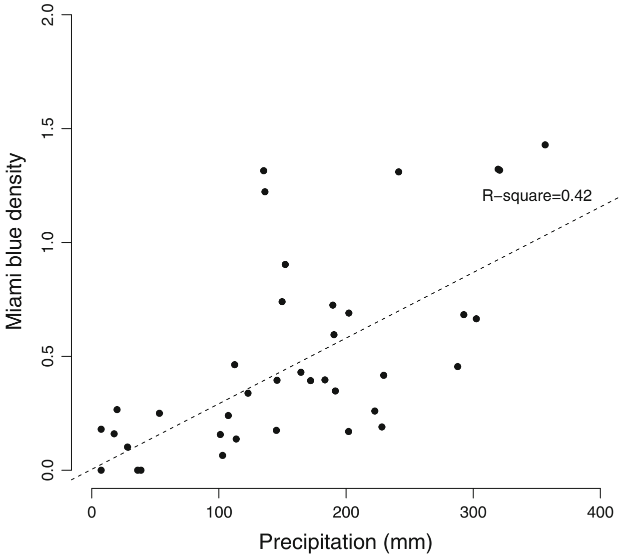


Fig. 5 Relationship between precipitation and butterfly density. X-axis represents the cumulative precipitation over 48-day period ending 28 days before survey date

We made one key adjustment to traditional point transect distance sampling methods to ensure we did not violate the assumption that butterflies are detected at their initial location: we further developed methods to determine the proper survey duration. Our analysis of the rate at which we accumulated detections validated our use of data from a 10 s survey for our density estimates, the optimal duration for Miami blue butterflies. This is much shorter than the 5-min survey recommended for birds (Rosenstock et al. 2002), although surveys as short as 2 or 3 min have been recommended for particularly active bird species (Cimprich 2009; Peak 2011). Short survey duration is necessary for Miami blues because they are most often detected on the wing and move toward and away from the observer rapidly. Additional methods could be used to validate survey duration. With knowledge of movement rates, one could simulate the rate at which butterflies move into and out of the estimated detection radius for a particular species. Repeat sampling at sites could also help determine survey duration by allowing for estimation of butterfly availability. Future attempts to apply point transects to insects that behave differently than Miami blues will need to evaluate the survey duration that best captures realistic densities of the target species (Lee and Marsden 2008).

In an ideal survey scenario, point locations would either be randomly distributed throughout the butterfly's habitat, or stratified in a predefined grid (Buckland et al. 2001). This would allow for unbiased estimation of butterfly density. In the impenetrable blackbead thicket characteristic of Miami blue habitat, navigating to random or stratified point locations is not feasible. Because of this limitation, we located survey points randomly on the edge of blackbead habitat. This decision has the potential to bias our density estimates either high or low if butterflies are attracted to, or avoid the blackbead edge.

A number of studies have examined butterfly behavior and density with respect to habitat edges (Haddad 1999; Haddad and Baum 1999; Ries and Debinski 2001; Schultz and Crone 2001; Ross et al. 2005; Schultz et al. 2012). In these cases, butterfly densities are greatest in the center of a habitat patch (Haddad and Baum 1999) and butterflies avoid patch edges from as far as 20 m away (Ries and Debinski 2001; Schultz and Crone 2001; Ross et al. 2005; Schultz et al. 2012). If Miami blues follow this established pattern, then we consistently underestimate Miami blue density. Previously studied landscapes, however, are very different from Miami blue habitat. Butterfly habitat in previous edge studies is generally open compared with the surrounding matrix (except Schultz and Crone 2001, where habitat and matrix are both open grassland), nectar and host plant resources co-occur, and resources are unavailable in adjacent habitat. Under these conditions, butterflies are attracted to openings, and avoid forests (Haddad and Baum 1999; Ries and Debinski 2001; Ross et al. 2005; Schultz et al. 2012). The structure of Miami blue habitat, however, is not open and grassy, but dense and shrubby, and nectar resources are not always available within host plant patches. Instead, many flowering plants occur in the adjacent, open, coastal prairie community. When important resources are located within adjacent habitat types, butterflies are less likely to respond to structural habitat boundaries (Schultz et al. 2012) and may even be attracted to the edge (Ries et al. 2004). Under this scenario, we would consistently overestimate Miami blue densities by only surveying along the blackbead edge. We surveyed a limited number of points within blackbead, and recorded individual Miami blues that we stumbled upon while mapping blackbead. We encountered a number of butterflies within the center and back edge (adjacent to mangrove) of the blackbead, even during periods of low abundance. These observations confirm that butterflies are at least present throughout blackbead, and suggest it is unlikely that we are grossly over or underestimating Miami blue density.

Another aspect of our design that has the possibility of biasing density estimates is our decision to orient our survey semi-circles perpendicular to the blackbead edge. If Miami blues are mainly detected on the blackbead side of the semi-circle, we are consistently under-estimating butterfly density. We did not collect data on the habitat type in which we detected butterflies, but future monitoring efforts could. These data would allow us to determine the proportion of butterflies detected in blackbead versus beach dune and refine our density estimates further.

The density and abundance estimates we present are an index of population size, the number of adult individuals estimated to be flying on a given day; they are not total population size. This number will be sufficient to track the size and trends of the Miami blue population over time. The true population size, however, would account for all adults that emerge over a flight period within one generation. Total butterfly population size is generally estimated by combining daily counts across a flight season (Mattoni et al. 2001). This approach works in temperate regions for butterflies with distinct flight periods, each representing a single generation. For Miami blues, we cannot simply sum counts because of the possibility of overlapping generations. This fact makes it nearly impossible to estimate total population size without estimating demographic parameters such as daily survival and generation time (Williams et al. 2002). Some estimates of both daily survival rates and generation time exist for Miami blue in captivity (J. Daniels, personal communication), but these estimates do not represent the seasonal or temporal variability that is expected for the Key West Refuge population. Given the variability we observed with respect to inter- and intra-annual phenology, estimating these demographic parameters in the field would be necessary to calculate accurate estimates of total population size.

Implications for an imperiled butterfly

Our estimates of peak Miami blue daily density are the first for the Key West Refuge. Peak daily densities of 500–600 butterflies per hectare are much higher than previously expected for Miami blues. In the winter of 2006/2007, when the butterflies were initially discovered in Key West refuge, Cannon et al. (2010) observed peak daily counts of 441 and 521 Miami blues on Boca Grande and Main Beach, respectively. Since those initial reports of abundances in the hundreds, surveys have been sporadic and failed to produce a daily count higher than 14 Miami blue butterflies (Daniels 2010). Rather than indicating a dramatic loss of butterflies, these surveys more likely reflect the difficulty of targeting surveys around periods of high adult abundance. By surveying continuously throughout the year, we captured time periods of both low and high Miami blue densities.

Peak Miami blue densities in 2012 and 2013 were similar, but intra-annual fluctuations were large (Fig. 2). It is unknown if low adult density translates to low overall population size or simply low *adult* abundance, therefore, low adult densities are concerning for management of Miami blues. The observed swings from low to high densities could result from erratic boom and bust population cycles not uncommon among insects. Our surveys, however, detected rapid increases from low adult density to high in just a week, instead of the gradual increase across multiple generations expected in the boom and bust scenario. A better explanation for the observed fluctuation in adult population size is that periods of low (or no) adult abundance represent times when immature Miami blue development rate is dramatically reduced, likely due to environmental conditions. This scenario fits with the precipitation model we developed (Fig. 5). Likely, early or mid-instar Miami blue larvae diapause during droughts as their blackbead host desiccates and becomes less palatable. Precipitation then initiates new growth of blackbead and larvae emerge from diapause, complete development, and fly as adult butterflies 4–6 weeks later. This period of larval dormancy explains the lag we observed between increased precipitation and peak butterfly abundance. Because we almost always saw at least one Miami blue, there is likely a high amount of variation in when, and for how long, an individual diapauses as well as how long an individual butterfly lives.

Despite high densities, the global distribution of Miami blues remains small and limited to low-lying islands where sea level rise will undoubtedly accelerate habitat loss over the next few decades. Another, perhaps more imminent, climate change related threat is the potential for changes in precipitation patterns. Because of their proximity to the Caribbean, the Florida Keys are likely to see decreases in precipitation by the end of the century (Collins et al. 2013). It is also likely that the contrast between wet and dry seasons will increase (Collins et al. 2013), potentially leading to much drier winters in the Florida Keys. If Miami blue larvae diapause during dry periods, prolonged drought could have negative effects on the population through reduction in adult size (Hahn and Denlinger 2007) which could result in reduced fecundity (Trager and Daniels 2011). Extended diapause also has the possibility of increasing the butterfly's vulnerability to predation and parasitism. In cold climates, parasitoids and invertebrate predators often diapause simultaneously with their prey (Hahn and Denlinger 2007). This is unlikely to be the case in the warm Florida Keys, where diapausing larvae are likely susceptible to a number of unknown invertebrate assassins.

Using point transect distance sampling to estimate insect density is a reasonable option for species whose habitat or life history traits make traditional methods impossible to implement. Still, careful attention must be paid to assure that assumptions of the method can be met. The need for 40–60 detections to accurately fit a detection function to the data may still not make point transects a feasible option for rare species that exist at extremely low densities. For these species, pooling data across space and/or time could allow for the development of a "global" detection function, as long as it is unlikely that detection probability varies in space and/or time. The double observer approach for estimating detection probability from points (Nichols et al. 2000) could also be applied to low-density butterflies. For some very rare species that are logistically challenging to monitor, land managers may have to rely on count index methods, or shift focus to tracking occupancy instead of abundance over time. Although Miami blue butterflies are rare globally, difficult to access, and occupy challenging habitat, they are still locally abundant and exist at densities high enough to allow for quantitative estimation of their population density.

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References

Boughton DA (2000) The dispersal system of a butterfly: a test of source-sink theory suggests the intermediate-scale hypothesis. Am Nat 156:131–144

Bried JT, Pellet J (2012) Optimal design of butterfly occupancy surveys and testing if occupancy converts to abundance for sparse populations. J Insect Conserv 16:489–499

Brown JA, Boyce MS (1998) Line transect sampling of Karner blue butterflies (*Lycaeides melissa samuelis*). Environ Ecol Stat 5:81–91

Buckland ST (2006) Point-transect surveys for songbirds: robust methodologies. Auk 123:345–357

Buckland ST, Anderson DR, Burnham KP et al (2001) Introduction to distance sampling: estimating abundance of biological populations. Oxford University Press, Oxford

Calhoun JV, Slotten JR, Salvato MH (2002) The rise and fall of tropical blues in Florida: *Cyclargus ammon* and *Cyclargus thomasi bethunebakeri*. Holarct Lepid 7:13–20

Cannon P, Wilmers T, Lyons K (2010) Discovery of the imperiled Miami blue butterfly (*Cyclargus Thomasi Bethunebakeri*) on islands in the Florida Keys National Wildlife Refuges, Monroe County. Southeast Nat 9:847–853

Cimprich DA (2009) Effect of count duration on abundance estimates of black-capped Vireos. J Field Ornithol 80:94–100

Collins M, Knutti R, Arblaster J, Dufresne JL, Fichefet T, Friedlingstein P, Gao X, Gutowski WJ, Johns T, Krinner G, Shongwe M, Tebaldi C, Weaver AJ, Wehner M (2013) Long-term climate change: projections, commitments and irreversibility. In: Stocker TF, Qin D, Plattner G-K, Tignor M, Allen SK, Boschung J, Nauels A, Xia Y, Bex V, Midgley PM (eds) Climate change 2013: the physical science basis contribution of working group i to the fifth assessment report of the intergovernmental panel on climate change. Cambridge University Press, Cambridge

Cordero-Rivera A, Perez FE, Andres JA (2002) The effect of handling damage, mobility, body size, and fluctuating asymmetry on lifetime mating success of *Ischnura graellsii* (rambur) (Zygoptera: Coenagrionidae). Odonatologica 31:117–128

Daniels JC (2010) Conservation and field surveys of the endangered Miami blue butterfly (*Cyclargus thomasi bethunebakeri*) (Lepidoptera: Lycaenidae) report #3. Submitted to United States Fish and Wildlife Service: Florida Keys National Wildlife Refuges

Daubenmire R (1959) A canopy-coverage method of vegetational analysis. Northwest Sci 33:43–64

Dover JW, Sparks TH, Greatorex-Davies JN (1997) The importance of shelter for butterflies in open landscapes. J Insect Conserv 1:89–97

Emmel TC, Daniels JC (2008) Miami blue butterfly conservation interim annual report. Period covered: January 2007-August 31, 2008. Submitted to United States Fish and Wildlife Service: South Florida Ecological Services Office

FNAI (2010) Guide to the natural communities of Florida, 2010th edn. Florida Natural Areas Inventory, Tallahassee

Haddad NM (1999) Corridor use predicted from behaviors at habitat boundaries. Am Nat 153:215–227

Haddad NM, Baum KA (1999) An experimental test of corridor effects on butterfly densities. Ecol Appl 9:623–633

Haddad NM, Hudgens B, Damiani C et al (2008) Determining optimal population monitoring for rare butterflies. Conserv Biol 22:929–940

Hahn DA, Denlinger DL (2007) Meeting the energetic demands of insect diapause: nutrient storage and utilization. J Insect Physiol 53:760–773

Hamm CA (2013) Estimating abundance of the federally endangered Mitchell's satyr butterfly using hierarchical distance sampling. Insect Conserv Divers 6:619–626

Henry EH, Haddad NM, Wilson JW (2012) Miami blue butterfly monitoring interim report covering September 2010–May 2012. 14. Submitted to United States Fish and Wildlife Service: Florida Keys National Wildlife Refuges

Isaac NJB, Cruickshanks KL, Weddle AM et al (2011) Distance sampling and the challenge of monitoring butterfly populations. Methods Ecol Evol 2:585–594

Kuefler D, Haddad NM, Hall S et al (2008) Distribution, population structure and habitat use of the endangered Saint Francis Satyr butterfly, *Neonympha mitchellii francisci*. Am Midl Nat 159:298–320

Lee DC, Marsden SJ (2008) Adjusting count period strategies to improve the accuracy of forest bird abundance estimates from point transect distance sampling surveys. Ibis 150:315–325

Mackenzie DI, Nichols JD, Royle JA, Pollock KH, Bailey LL, Hines JE (2006) Occupancy estimation and modeling: inferring patterns and dynamics of species occurrence. Academic Press, London

Mattoni R, Longcore T, Zonneveld C, Novotny V (2001) Analysis of transect counts to monitor population size in endangered insects the case of the El Segundo blue butterfly, *Euphilotes bernardino allyni*. J Insect Conserv 5:197–206

Moranz RA, Fuhlendorf SD, Engle DM (2014) Making sense of a prairie butterfly paradox: the effects of grazing, time since fire, and sampling period on regal fritillary abundance. Biol Conserv

Murphy DD (1987) Are we studying our endangered butterflies to death? J Res Lepid 26:236–239

Nichols JD, Hines JE, Sauer JR et al (2000) A double-observer approach for estimating detection probability and abundance from point counts. Auk 117:393–408

Peak RG (2011) A field test of the distance sampling method using Golden-cheeked Warblers. J Field Ornithol 82:311–319

Pocewicz A, Morgan P, Eigenbrode SD (2009) Local and landscape effects on butterfly density in northern Idaho grasslands and forests. J Insect Conserv 13:593–601

Pollard E (1977) A method for assessing changes in the abundance of butterflies. Biol Conserv 12:115–134

Powell AFLA, Busby WH, Kindscher K (2006) Status of the regal fritillary (*Speyeria idalia*) and effects of fire management on its abundance in northeastern Kansas, USA. J Insect Conserv 11:299–308

Ries L, Debinski DM (2001) Butterfly responses to habitat edges in the highly fragmented prairies of Central Iowa. J Anim Ecol 70:840–852

Ries L, Fletcher RJ Jr, Battin J, Sisk TD (2004) Ecological responses to habitat edges: mechanisms, models, and variability explained. Annu Rev Ecol Evol Syst 35:491–522

Rosenstock SS, Anderson DR, Giesen KM et al (2002) Landbird counting techniques: current practices and an alternative. Auk 119:46–53

Ross JA, Matter SF, Roland J (2005) Edge avoidance and movement of the butterfly *Parnassius smintheus* in matrix and non-matrix habitat. Landsc Ecol 20:127–135

Saarinen EV, Daniels JC (2006) Miami blue butterfly larvae (Lepidoptera: Lycaenidae) and ants (Hymeoptera: Formicidae): new information on the symbionts of an endangered taxon. Fla Entomol 89:69–74

Saarinen EV, Daniels JC (2012) Using museum specimens to assess historical distribution and genetic diversity in an endangered butterfly. Anim Biol 62:337–350

Saarinen EV, Daniels JC, Maruniak JE (2009) Development and characterization of polymorphic microsatellite loci in the endangered Miami blue butterfly (*Cyclargus thomasi bethunebakeri*). Mol Ecol Resour 9:242–244

Schultz C, Crone E (2001) Edge-mediated dispersal behavior in a prairie butterfly. Ecology 82:1879–1892

Schultz CB, Franco AMA, Crone EE (2012) Response of butterflies to structural and resource boundaries. J Anim Ecol 81:724–734

Simons TR, Alldredge MW, Pollock KH et al (2007) Experimental analysis of the auditory detection process on avian point counts. Auk 124:986–999

Sparrow HR, Sisk TD, Ehrlich PR, Murphy DD (1994) Techniques and guidelines for monitoring neotropical butterflies. Conserv Biol 8:800–809

Thomas JA (1983) A quick method for estimating butterfly numbers during surveys. Biol Conserv 27:195–211

Thomas L, Buckland ST, Rexstad EA et al (2010) Distance software: design and analysis of distance sampling surveys for estimating population size. J Appl Ecol 47:5–14

Trager MD, Daniels JC (2011) Size effects on mating and egg production in the Miami blue butterfly. J Insect Behav 24:34–43

USFWS (2012) Endangered and threatened wildlife and plants; listing of the Miami blue butterfly as endangered throughout its range;

listing of the cassius blue, ceranus blue, and nickerbean blue butterflies as threatened due to similarity of appearance to the Miami blue butterfly in coastal south and central Florida. 77:20948–20986

van Strien AJ, van Swaay CAM, Termaat T (2013) Opportunistic citizen science data of animal species produce reliable estimates of distribution trends if analysed with occupancy models. J Appl Ecol 50:1450–1458

Wikström L, Milberg P, Bergman K-O (2008) Monitoring of butterflies in semi-natural grasslands: diurnal variation and weather effects. J Insect Conserv 13:203–211

Williams B, Nichols JD, Conroy MJ (2002) Analysis and management of animal populations. Academic, London

Wolda H (1988) Insect seasonality: Why? Annu Rev Ecol Syst 19:1–18