Journal of Insect Conservation Survival of eggs to third instar of late-summer and fall-breeding monarch butterflies (Danaus plexippus) and queen butterflies (Danaus gilippus) in north Texas

Alyx Scott
Houston Zoo
Kelsey Contreras
University of Texas at Arlington
Misty Stevenson
Dallas Arboretum and Botanical Garden
Kalynn L. Hudman
Texas A&M University – Commerce
Jeffrey G. Kopachena (Jeff.Kopachena@tamuc.edu)
Texas A&M University – Commerce

Research Article

Keywords: Monarch Butterfly, Danaus plexippus, Queen Butterfly, Danaus gilippus, fall survival, phenology, Texas

Posted Date: October 11th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1739437/v1

Abstract

Introduction: Eastern migratory monarch butterflies (*Danaus plexippus*) have declined over 80% in recent years, but little is known about fall reproduction in the southern U.S. where monarchs may compete with queen butterflies (*Danaus gilippus*).

Aims/Methods: We provide data on the survival to third instar, associated arthropods, and phenology of fall breeding monarchs and queens in Texas.

Results: Monarch and queen survival was high, but varied among years. Oleander aphids (*Aphis nerii*), jumping spiders (Salticidae), and red imported fire ants (*Solenopsis invicta*) had minor negative effects on survival. The abundance of monarchs and queens on the study site peaked three to four weeks before the main passage of monarchs in the area. Queens had similar phenology and exhibited a migratory pattern similar to monarchs but on a smaller scale.

Discussion: Survival of fall monarchs is high and likely to be important for winter roost recruitment. Fall egg survival was not greatly affected by any particular arthropod taxon, but may be affected by precipitation. Fall reproduction is a response to available host plants and likely adaptive. The timing of oviposition enables pupae to eclose in time for successful migration to winter roosts.

Implications for Insect Conservation: Management of *Asclepias viridis*and other native milkweeds to facilitate fall reproduction is important for the recovery of monarchs because it buffers variable productivity occurring further north and contributes significantly to overwintering populations. Management should use mowing and burn schedules that promote high quality host plants. Populations of queens should be monitored for their potential to compete with monarchs especially in response to the potential impacts of parasite resistance and climate change.

Introduction

The monarch butterfly (*Danaus plexippus*) is an iconic species distributed across much of North America (Gustafsson et al. 2015). Based on winter roost census data, the eastern migratory population of this species has shown alarming declines in recent years (Thogmartin et al. 2017; Brower et al. 2018) with some models projecting extinction thresholds within the next few decades (Flockhart et al. 2015; Semmens et al. 2016; Oberhauser et al. 2017; Voorhies et al. 2019; Nail 2020). In response, the Monarch Joint Venture established the Monarch Conservation Implementation Plan (Monarch Joint Venture 2021) focused on enhancing breeding habitat, migratory habitat, and protecting overwintering sites. A major emphasis of the Monarch Conservation Implementation Plan is to restore and enhance pollinator habitat and to increase milkweed host-plant populations. The efficacy of these strategies depends on underlying spatial and temporal assumptions about monarch survival. However, there are important gaps in this data. This is particularly true with regard to the reproductive output of fall-breeding individuals in the southern U.S. (Flockhart et al. 2013).

There are small populations of monarchs in Florida, on Caribbean islands, and along the extreme margins of the Gulf of Mexico that are permanent residents (Howard et al. 2010; Dockx 2012; Zanden et al. 2018) and in some locations, particularly along the Gulf of Mexico, some of these individuals either simply overwinter or breed through the winter (Howard et al. 2010). In Florida and most likely in other locations with resident populations, there is also mixing between resident individuals and migratory individuals (Knight and Brower 2009; Zanden et al. 2018). Aside from these smaller populations, almost all of the eastern migratory population of monarch butterflies spend their winter in a few roost sites in central Mexico (Vidal and Rendón-Salinas 2014). In the spring, these butterflies migrate north and re-colonize most of North America east of the Rocky Mountains through a series of successive generations (Flockhart et al. 2013). Traditionally, it has been assumed that the last and most northern generation enters reproductive diapause and migrates south to the overwintering sites in Mexico (Malcolm et al. 1993). These individuals then migrate north in spring to begin the cycle anew.

However, many monarchs migrating through southern latitudes in late summer or early fall either come out of reproductive diapause or have never entered diapause (Batalden and Oberhauser 2015; Majewska and Altizer 2019). These individuals lay eggs in the southern U.S., thereby resulting in an additional generation (Calvert 1999; Borland et al. 2004; Prysby and Oberhauser 2004; Flockhart et al. 2013; Brym et al. 2018). There is considerable evidence that fall reproduction may provide a substantial addition to the overwintering population in Mexico (Flockhart et al. 2013). Stable isotope data demonstrate that about 11% of monarch butterflies at overwintering sites in Mexico originate from south-central portions of the U.S. and, in some years, up to 30% of the wintering population may originate from areas of Texas, Oklahoma, Louisiana, and Arkansas (Flockhart et al. 2017). Importantly, fall breeding appears to augment the overwintering population in years when productivity in the U.S. Midwest is poor. These data emphasize the need to focus monarch conservation across the entire breeding distribution in appropriate places at appropriate times (Flockhart et al. 2017). Current monarch breeding habitat conservation focuses on spring and summer reproduction and largely neglects fall breeding in the south because fall breeding is poorly understood, especially in the critically important migratory corridor in Oklahoma and Texas.

There is no detailed information on the survival of fall-breeding monarch eggs and larvae along the main corridor in the southern U.S. or on the ecological context of this generation and its associated host-plant arthropods. The available information places focus on milkweed availability along with egg and larval abundance (Calvert 1999; Prysby and Oberhauser 2004; Baum and Sharber 2012; Baum and Mueller 2015) or emphasizes the potential impact of exotic milkweed on enhanced parasite exposure and stimulation of reproductive behavior among migrants (Batalden and Oberhauser 2015; Satterfield et al. 2015; Majewska and Altizer 2019; Clement and Crawford 2020). One study demonstrated that field collected late instars show high levels of parasitism (Mueller and Baum 2014), but did not quantify overall survivorship of eggs to any particular stage of development. Knowledge of the survivorship of these fall-breeding monarchs is needed to determine what factors affect survival as well as to inform efforts that facilitate fall breeding as a strategy in the recovery of eastern migratory monarchs. In this paper we provide the first detailed information on fall-breeding monarch egg survival to the third instar at a site in northeast Texas. In addition, we provide information on the phenology of adult and larval site occupancy in order to put fall reproduction into a temporal context.

The focus of our study was fall reproduction in monarch butterflies. However, it quickly became apparent that we also needed to document queen butterflies (*Danaus gilippus*) which breed in the same area in fall, share the same host plants, and are potential competitors (Brower 1962; Brym et al. 2020). Furthermore, while the eastern subspecies of the queen is well studied in Florida (Brower 1961, 1962; McLaughlin and Myers 1970; Ritland 1991a, b; Moranz and Brower 1998), and there is some literature on the western subspecies of queens in the Mojave and Sonoran Deserts (Sakai 1993; Grodsky et al. 2020; Saul-Gershenz et al. 2020), there is almost no literature on the biology of the queen butterfly in Texas. What little is known indicates that queen butterflies in Texas migrate back and forth into Mexico (Einem 2003; Hobson et al. 2021). This migratory tendency coupled with the general potential for tropical butterflies to expand northward in response to warming climates (Kocsis 2011) makes it likely that queen butterflies are also expanding their distribution northward (Moskowitz 2003). Monarchs and queens appear to exhibit density-dependent competition (Brower 1961, 1962), but monarchs are more vulnerable to protozoan ectoparasites (Barriga et al. 2016) and may be less adaptable to climate change (York and Oberhauser 2002; Zipkin et al. 2012; Zylstra et al. 2021). If so, then the competitive dynamic between monarchs and queens could favor queens under future climate scenarios. This makes it important to monitor queen populations as they expand into the U.S. Because queens are an important aspect of the ecology of fall reproduction by monarchs, we felt it necessary to provide data on the reproductive success, abundance, and phenology of queens at our study site.

Lastly, we wanted to document the ecological context of fall reproduction for both monarchs and queens relative to the arthropod community associated with milkweed host plants. The aim was to see how these arthropods might affect egg and larval survival. We pay added attention to the possible effects of red imported fire ants (*Solenopsis invicta*)

(hereinafter referred to as fire ants) as this species has been implicated as a major predator of monarchs in the southern U.S. (Calvert 1996, 2004).

Materials And Methods

Study Site

The study was conducted on a 28-hectare old-field on the outskirts of the city of Sulphur Springs, Hopkins County, Texas (33˚07’49.1” N, 95˚39’03.3” W). The site, owned by the Sulphur Springs Economic Development Corporation (SSEDC), was vacant land typically mowed in spring, summer, and fall, in order to suppress woody vegetation and to harvest hay. During the two years of our study, the field was mowed in early May, mid-July, and after we had finished collecting data, at the end of October in 2017 and the middle of November in 2018. Historically, the land had been used for crop cultivation but, more recently, and prior to acquisition by the SSEDC, the land was converted for hay production and planted with Bahia grass (*Paspalum notatum*) and Bermuda grass (*Cynodon dactylon*). The site had relatively low plant diversity and was largely dominated by grasses. However, at least ten years of minimal management after the land was acquired by the SSEDC led to the invasion of this field by a variety of other grasses and forbs. In particular, the field contained a high density of Green Milkweed (*Asclepias viridis*), the only species of milkweed on the study site. On 25 September 2017, an analysis of 35 randomly placed 50 m^2 circular quadrats found the density of the milkweed plants to be 0.21 $\pm$ 1.88 plants/m^2 or 2069 $\pm$ 373.8 plants/ha.

Data Collection

Data were collected daily from 15 August 2017 through 26 October 2017 and from 15 August 2018 through 3 November 2018. Field seasons ended when eggs could no longer be found and when the last of our focal individuals had either disappeared or reached the third instar. An exception to daily monitoring occurred in early September of 2018 when a human body was found on the study site. As a result, from 2 September through 4 September 2018, we were unable to access the study site due to the resulting criminal investigation.

Eggs were found by either watching females lay eggs or by searching potential host plants for eggs. When an egg was found, the leaf, stem, or flower on which the egg was attached was marked with a non-toxic ink marker. A white numbered flag was placed adjacent to the plant so that it could be located in the future. Following the method used in other focal individual studies (Prysby 2004; De Anda and Oberhauser 2015; Stevenson et al. 2021), each egg was visited daily between 08:00h and 14:00h CDST to monitor its status. Monarch larvae sometimes temporarily leave the host plant (Rawlins and Lederhouse 1981; Borkin 1982) and early instars can be difficult to locate even if present on the host plant (Prysby and Oberhauser 2004). Therefore, when an egg or larva was missing from a host plant, the plant was revisited for three subsequent days to ensure that the larva had not simply been off the plant or overlooked. As in other studies on monarch survival (Zalucki and Kitching 1982; Zalucki et al. 1990), we considered eggs and larvae that were missing to be mortalities and the date of the mortality was recorded as the first day the egg or larva was missing from the host plant. Monarch larvae begin to emigrate from the host plant once they reach the third instar (De Anda and Oberhauser 2015; Fisher et al. 2020) and at that point we could not distinguish mortality from emigration. For this reason, we terminated observations of an individual once it was missing or reached the third instar. Therefore, our estimates of survival are limited to survival to the third instar.

It was not possible to distinguish the eggs of monarchs from the eggs of queens in the field (Calvert 1999; Batalden and Oberhauser 2015; Brym et al. 2018; Scott 2019) so we initially had to combine all of the eggs for analyses of egg survival to the first instar. First instar queens can be distinguished from first instar monarchs by the presence of a pair of black dots corresponding to the third pair of tubercles present in queen butterfly larvae. This enabled us to document the survivorship from first to third instar separately for each species.

During daily visits to the host plants, all arthropods on the plant were documented. To do so, we approached each plant carefully and recorded any volant insects during that approach. More sedentary arthropods were noted upon closer inspection of the plant and while checking on the status of the monarch or queen egg or larva. In order to minimize disturbing the arthropods, we could not collect them. This technique clearly has limitations. First, it only provides a snapshot of the arthropods on a host plant. Second, many arthropods require close inspection of genitalia or other fine structures to make precise identification. Though some species were clearly identifiable, many arthropods could only be identified to family. Consequently, our interpretation of these data is highly generalized and, though the data provide information on arthropod community composition, our ability to correlate particular arthropods with survival is limited.

Fire ants have been implicated as having catastrophic effects on monarch eggs and larvae in Texas (Calvert 1996, 2004) and this merited closer evaluations of the potential effects of fire ants. In addition to noting fire ants on the host plants, and following the method of Hudman (2018), we quantified the density of fire ants adjacent to the host plant by counting all fire ant mounds within 4m of the host plant on the day that an egg was first located. The purpose of this data was to determine if monarch or queen survival was correlated with fire ant mound density adjacent to the host plants.

During the course of checking the status of eggs and larvae, and while searching for new eggs on new host plants, we kept track of the number of non-focal larvae found on non-focal host plants and on the numbers of adult monarch and queen butterflies observed on the study site. We also kept track of how many plants were examined to provide a crude measure of search effort. We used these data to generate a measure of the phenology of monarchs and queen adults and larvae on our study area. For adults we were able to compare our data on monarch phenology with data provided by Journey North (Sheehan and Weber-Grullon 2021). To do so, we downloaded data from the Journey North website and included only fall monarch sightings recorded between 15 August and 15 November for both 2017 and 2018. We restricted the data to observations that occurred between 34^{o} N and 32^{o} N and between 94^{o} W and 98^{o} W. This is an area that encompassed a region 111 km north or south of the study site and 186 km east or west of the study site. We compared our data on the phenology of adult queens with that provided by Butterflies and Moths of North America (BAMONA)(Lotts and Naberhaus 2021). This data was much sparser than the Journey North Data. For that reason, and because the overall seasonal phenology of queens is so poorly documented in Texas, we evaluated BAMONA data that spanned the entire annual cycle for 20 years from 1 January 2000 through 31 December 2020. These data were then stratified into three broad geographic regions in Texas, all east of 100^{o} W: South Texas (Mexican border and Gulf of Mexico to 28^{o}N), Central Texas (28^{o} N to 31^{o} N, 96^{o} W to 100^{o} W), North Texas (31^{o} N to Oklahoma border and 100^{o} W east to Louisiana border).

Weather conditions varied dramatically among the two years of the study. To quantify these differences, daily weather data on precipitation and temperature were obtained from a National Oceanic and Atmospheric Administration (NOAA) weather station located 2.3 km from the study site. All statistical analyses were conducted using SAS® Studio 3.8 software. For frequency data (number survived or died, number of fire ant mounds) we used Chi-square Contingency Tables. When some cells had expected values of less than five, Fisher's Exact Tests were used. Logistic Regression was used to analyze the effect of arthropods on egg and larval survival. This analysis used a step-wise procedure to isolate the best subset of models. AICc was then used to select the best model from that subset. For data on site occupancy by adults and non-focal larvae, two-sample t-tests were used to compare among species or years. Folded F-tests were used to determine if variances were equal. Where variances were unequal, Satterthwaite's method for unequal variances was used to estimate the test statistic, otherwise the pooled variance estimate was used to estimate the test statistic. For all statistical analyses we used p-values to assess the level of confidence associated with effect sizes (Murtaugh 2014).

Results

Weather Conditions

We present these data because markedly different weather conditions between the two years of the study may have had important impacts on the results. In particular, drought conditions during 2017 resulted in obvious water stress to the host plants that was not evident during the much wetter year of 2018. These differences likely impacted egg and larval survival.

Temperatures did not differ markedly from normal for either year, though August of 2018 was somewhat hotter than normal (Table 1). For our site, the average first day of frost is November 11. However, in 2017 the temperature fell below freezing on the mornings of 28 October and 29 October, two days after the last two eggs disappeared, four days after the last monarch larva reached the third instar, and 13 days after the last queen reached the third instar. Low temperature on each of these mornings was -0.6^{o}C. In 2018, temperatures remained above freezing until 10 November when the morning temperature was -1.7^{o} C. This was seven days after the last egg was missing, 13 days after the last monarch reached the third instar, and 14 days after last queen reached the third instar.

Precipitation differed markedly among years (Table 1). The 30.3 cm of precipitation for August 2017 is misleading because that total reflects a single precipitation event on 13 August when 20.83 cm of rain fell. Otherwise, the fall of 2017 was extremely dry. Only 0.66 cm of rain fell between 19 August and 2 October and there were only nine days of precipitation over the course of the study. These dry conditions led to observable wilting and loss of condition of many plants, including some milkweed host plants.

In contrast, the fall of 2018 was extremely wet (Table 1). Precipitation in September was 167% above normal and precipitation in October was 43% above normal. In 2018, it rained on 28 of the 81 days of the study. Though there was ponding of water on many parts of the study area, there were no cases in which a mortality could be linked to a host plant immersed in water. The increased precipitation in 2018 resulted in a noticeably better condition of the host plants.

Survival of eggs and larvae

Over the two years of the study, 463 eggs were found on 414 plants; 231 eggs in 2017 and 232 eggs in 2018. The peak period of egg-laying occurred in September. Despite marked differences in weather conditions among the two years, the density of eggs per plant checked varied little among the two years of the study. In September of 2017, an average of 36 potential host plants had to be examined to find one egg. In September of 2018, an average of 31 potential host plants had to be searched to find one egg. Because there was no way to distinguish monarch eggs from queen eggs in our population (Scott 2019), hatching success included both monarchs and queens. Across both years, 51% of eggs survived to hatch. However, hatching success differed among years (Fig. 1). In 2017, 44.6% of eggs hatched and the daily survival rate was 83%. In the wetter year of 2018, 67.2% of eggs hatched and the daily survival rate was 92%. In 2017, 11.7% of first instars survived to the third instar and in 2018, 26.3% of first instars survived to the third instar (Fig. 1). Overall, 5.2% of eggs survived to the third instar in 2017, whereas in 2018, 17.7% of eggs survived to third instar.

First instar monarchs and queens are extremely small and, in some cases, owing location on the plant, light conditions, and the need to avoid disturbing the larvae, it was not possible to immediately identify the larvae. Some of these unidentified individuals disappeared before they could be identified to species. As a result, 37 of the103 eggs that hatched in 2017 and 12 the 153 eggs that hatched in 2018 succumbed before they could be identified. Thus, we identified 56 first instar monarchs and 10 first instar queens in 2017 and 73 first instar monarchs and 68 first instar queens in 2018.

Among first instars that were identified, 19.6% of monarchs and 10% of queens survived to the third instar in 2017 whereas, in 2018, 20.5% of monarchs and 38.2% of queens survived to the third instar (Fig. 2). These values are inflated because they do not include individuals that hatched but were not identified prior to their disappearance. However, the proportion of adults of each species observed in the field was highly concordant with the number of non-focal larvae of each species observed in the field (see phenology of monarchs and queens below). Based on the assumption that unidentified mortalities among first instars were also concordant with these proportions, we further refined our estimates of

survival for each species. In 2017, 80% of adults and non-focal larvae were monarchs, so we assigned 80% (n = 30) of the 37 unidentified larvae as monarchs and the remainder (n = 7) as queens. In 2018, 50% of adults and non-focal larvae were monarchs, so half of the 12 unidentified larvae (n = 6) were assigned as monarchs and the other half as queens. These values were used to estimate the survival of each species from hatching to third instar (Fig. 2). Based on these estimates, monarch survivorship from hatching to third instar was 12.8% in 2017, much higher than the 5.9% survivorship estimated for queens. In 2018, monarch survival from hatching to third instar was estimated to be 19%, much lower than the 36.5% survivorship estimated for queens (Fig. 2). Based on these estimates, in 2017 survival from egg to third instar was 5.7% for monarchs and 2.6% for queens. In 2018, survival from egg to third instar was 12.8% for monarchs and 24.5% for queens.

Host Plant Arthropods

Other arthropods on the host plants were documented to understand the frequency and abundance of predatory and non-predatory arthropods associated with monarch and queen eggs and larvae. Small sample sizes necessitated that these analyses combine data for both monarchs and queens. We also had to correct inherent biases in the data. Eggs that survived to the third instar were monitored longer than eggs that did not survive and this difference inflates the observed number and abundance arthropods on the host plants. To correct this, we removed from the analyses all eggs that were monitored for fewer than eight days. This left 105 eggs on 100 host plants; of these, 38 survived to the third instar and 67 died. The mean and variance for the length of time that individuals were monitored was comparable for eggs that survived and those that died (F-Test for Equal Variances, F = 102, df = 66, 37, p = 0.9802; t-test for means, t = 0.44, df = 103, p = 0.6621).

Associated with the 105 eggs were 2,192 individuals of 40 different taxa (Appendix 1). Fourteen taxa were considered predatory whereas the remaining 26 taxa were primarily non-predatory. The most abundant arthropods were oleander aphids, mainly because these aphids formed large aggregations on some host plants. Because of this tendency to form mass aggregations, the actual frequency of oleander aphids was very low. Only 9 of the 105 eggs (8.6%) were associated with oleander aphids. Most other arthropod taxa had low abundance. The second most abundant taxon was large milkweed bugs, another aggregating species, in which only 35 individuals were found. On average, with the exclusion of oleander aphids, the average host plant only harbored 2.9 + 0.58 (SE) arthropods, of which 1.4 + 0.36 (SE) were predators and 1.6 + 0.36 (SE) were non-predators. The most frequent taxon, and the most frequent predator, was jumping spiders which were associated with about 14% of host plants (Appendix 1). Grasshoppers were the most frequent non-predatory arthropod, occurring on about 12% of host plants. Thirty-seven of the 40 taxa had frequencies of less than 10% and a third of the host plants had no arthropods documented on them at all.

Because frequencies and abundances were so low, it was difficult to isolate specific taxa that had a noticeable impact on monarch or queen survival. Furthermore, sparse data can lead to inflated parameter estimates when using logistic regression (Greenland et al. 2016). To avoid this, we reduced the number of variables by combining some taxa into broader, more inclusive categories. The categories used included jumping spiders, grasshoppers, fire ants, and oleander aphids. The remaining taxa were combined into two groups; all other predatory taxa and all other non-predatory taxa (see Appendix for classification of feeding modes). Using a stepwise variable selection procedure we generated four predictive models, none of which were particularly satisfactory (Table 2). The best model, based on corrected AIC, contained four arthropod groups: fire ants, jumping spiders, all other non-predatory arthropods, and oleander aphids. All of these arthropod groups, except all other non-predatory arthropods, were negatively associated with monarch survival. The largest effects were a negative impact of fire ants and jumping spiders on monarch and queen survival. However, high p-values suggest a low level of confidence in these parameter estimates (Table 2).

The results of the logistic regression were not surprising given the sparsity of the data and the fact that our methods do not account for arthropod presence when the plants were not under observation. However, other studies have implicated fire ants as having very strong negative impacts on monarch egg and larval survival (Calvert 1996, 2004), so we evaluated

whether survival was correlated with the number of fire ant mounds within 4m of each host plant. We used the data from 462 eggs for which fire ant mound density was measured.

On average there were 0.74 $\pm$ 0.05 (SE) fire ant mounds within 4m of the host plants. This equates to a density of 147 fire ant mounds per hectare though slightly more mounds were observed in 2018 than in 2017 (Kruskal – Wallis Test, Chi-square Approximation, X^2 = 3.84, df = 1, p = 0.0502) (Table 3). Since we could not differentiate monarch and queen eggs, we conducted separate analyses for all eggs and for those monarch and queen larvae that were identified at the first instar (Table 3). Though none of the comparisons had p-values greater than 0.05, a couple of trends are evident. In 2017, there were more than four times as many fire ant mounds adjacent to host plants with mortalities as compared to host plants without mortalities. For monarch larvae monitored in both years combined, there were almost twice as many fire ant mounds adjacent to host plants with mortalities as compared to host plants without mortalities. These trends were not evident for eggs in 2018 or for queen larvae in both years combined.

Phenology of Monarchs and Queens

Data were collected on site occupancy by monarch and queen adults and larvae while searching potential host plants for eggs. For these analyses, days when fewer than ten plants were searched were excluded to ensure sufficient effort was represented. The number of adults or non-focal larvae observed was divided by the number of plants searched to generate a crude value of individuals per unit effort.

There were many more adult monarchs observed in August of 2017 than were observed in August of 2018 (Fig 3). The average date of site occupancy was 6 September in 2017 and 15 September in 2018. However, dates in September when large flights of monarchs occurred were similar for both years though some flights in 2018 were much larger than any flights in 2017 (Fig. 3). In 2017 large flights of monarchs occurred on 10, 17 and 19 September. In 2018, there were large flights of monarchs on 11 and 20 September (Fig. 3). These dates are about three to four weeks earlier than the average dates for monarch observations recorded by Journey North (Sheehan and Weber-Grullon 2021) (Fig 4). The Journey North data yielded 416 observations on 76 days in 2017 and 453 observations on 80 days in 2018 and generated average dates for monarchs in our area as 7 October in 2017 and 15 October in 2018.

Adult queens showed a similar temporal pattern of site occupancy each year (Fig. 3). The average date of site occupancy for queens was 18 September in both years and large flights of queens occurred on 14 and 17 September of 2017 and on 11, 19, 20, and 22 September of 2018. Adult queens were far more common in 2018 than they were in 2017. Though monarchs outnumbered queens in 2017, by the end of September of 2018, adult queens slightly outnumbered adult monarchs. In 2017, 80% of adults observed on the study site were monarchs. In 2018, monarchs represented only 50% of the adults observed on the site. Because of large numbers of adult monarchs in August of 2017, the average date of site occupancy was twelve days earlier for monarchs than it was for queens. In 2018 the average date of site occupancy by monarchs was only three days earlier than it was for queens.

Queens are poorly studied in Texas and we wanted to know where the queens on our study site may have originated. Data on the annual phenology of queens for Texas were obtained from the Butterflies and Moths of North America (BAMONA) website (Lotts and Naberhaus 2021). Though sample sizes were small, there were clear geographic and temporal patterns to queen occupancy in the three regions of Texas analyzed (Fig. 5). In south Texas, queens were mostly observed from January through March and again in October and November. No queens were recorded in south Texas for the months of July through September. In central and north Texas, peaks occurred in July and October. In these two regions, there were no observations of queens from January through March (Fig. 5). These data are consistent with an annual colonization of Texas that starts in January in south Texas and along the gulf coast, continues northward in April, and peaks in July and October in central and north Texas. At our study site, adult queen abundance was higher in September than it was in October. However, the eggs laid by our queens in September would mature and eclose sometime in October and the large

numbers observed in the BAMONA database in October at our latitude might represent newly emerged adults from this generation.

Data on the phenology of monarch and queen non-focal larvae was combined for all instars observed while searching for new eggs on new host plants. In 2017, the phenology of monarch and queen non-focal larvae was similar to that of the adults (Fig. 6) and monarch larvae were much more common than queen larvae. There was a peak in both monarch and queen larvae in mid- to late September. In 2017, the average date for monarch larvae was 12 September, six days earlier than the average date for queen larvae. In 2018, the numbers of both monarch and queen larvae increased as the season progressed. In that year, the average date for monarch larvae was 28 September, two days earlier than for queen larvae. In 2017, 82% of larvae observed while checking potential host plants were monarchs. In 2018, monarchs represented 44% of the larvae observed in the field. These percentages are similar to the percentages of adults observed in the field and, like the adults, the average dates of monarch larvae on the study site were well in advance of the average passage date of adults recorded by Journey North (Sheehan and Weber-Grullon 2021).

Discussion

Our estimates of the survival of fall-breeding monarchs and queens from egg to third instar varied considerably among the two years of the study; for monarchs, success was over two times higher in 2018 than it was in 2017. For queens, success was almost ten times higher in 2018 than it was in 2017. We attribute these differences among years to the very dry conditions of 2017 and the much wetter conditions of 2018. In 2017, many host plants exhibited visible signs of water stress in the form of wilting, reduced growth, and die-back of some ramets. We suspect that these effects of precipitation on host plant quality had an important impact on monarch and queen survivorship. Clearly more data are needed on the effects of host plant quality on fall reproduction in monarchs and queens as has been suggested by others (Prysby and Oberhauser 2004; Majewska and Altizer 2019).

Despite differences among years, the survivorship of fall monarchs from egg to third instar was high. The 12.8% survivorship estimated for 2018 was comparable to high survivorship to third instar reported for spring generation monarchs in Texas (Stevenson et al. 2021) and in Florida (Cohen and Brower 1982; Zalucki and Brower 1992; Brower et al. 2018). Even in 2017, our estimated survivorship of 5.7% was higher than rates reported for other studies reporting survivorship to third instar using similar protocols in Minnesota (De Anda and Oberhauser 2015), Wisconsin (Borkin 1982; Prysby 2004), and Michigan (Haan and Landis 2019). These comparisons show that fall reproduction by monarchs may be as productive, and possibly more productive, than earlier generations further north. Though we feel that our study site represents a typical old-field in north Texas, it is important to note that monarch survival can vary considerably among sites within a geographic region. For example, a study on spring survival in north Texas and northwestern Louisiana found that survival from egg to third instar varied from 0% to 40% (Lynch and Martin 1993). Also, our study is limited to survival from egg to third instar. Though much evidence shows that survival of older instars is higher than that of younger instars (Zalucki and Kitching 1982; Lynch and Martin 1993; Zalucki et al. 2001; Oberhauser et al. 2001; Prysby and Oberhauser 2004), such observations are based on data on spring and summer generations. There may be high levels of parasitism in fall-generation monarchs in the south (Mueller and Baum 2014), so the assumption of higher survival of older instars needs further investigation for fall breeding monarchs. Data are needed, covering a broader geographic area, multiple land use types, and older instars to consolidate the assessment of fall survival of monarch eggs and larvae.

Arthropod predators are important determinants of survival for monarch eggs and larvae (Prysby 2004; Koch et al. 2005; De Anda and Oberhauser 2015; Oberhauser et al. 2015; McCoshum et al. 2016; Hermann et al. 2019; Myers 2019; Myers et al. 2019, 2020; Nestle et al. 2020; Baker and Potter 2020) and this applies to queens and other lepidoptera as well (Zalucki et al. 2002). The milkweed plants in our study site harbored a wide diversity of arthropods other than monarchs and queens. Of 40 taxa documented, 14 were predatory and 26 were milkweed herbivores, transients, and other primarily non-predatory arthropods. However, the number of arthropods per host plant was small, making it difficult to isolate specific

taxa of importance in affecting survival. Three taxa seemed potentially important: oleander aphids, jumping spiders, and fire ants. Aphids are known to attract predators onto host plants (Prysby 2004), but in our study aphids also had a direct effect on host plant quality by causing extensive tissue damage particularly to the more delicate shoots and younger leaves. This damage may directly affect monarch survival or force younger instars to leave the plant which, at ages prior to the third instar, would likely be fatal (Zalucki and Kitching 1982; Zalucki et al. 2002; De Anda and Oberhauser 2015). Some plants on the study site showed extremely high infestations of oleander aphids and none of the most heavily infested plants held monarch or queen eggs. Of the 9 eggs associated with oleander aphids, only one held an egg that survived to the third instar and that particular host plant held only 15 oleander aphids. It seems likely that the presence of intense infestations of oleander aphids could reduce the availability of suitable host plants.

Jumping spiders were the most frequent predators, though they occupied just under 15% of host plants. Collectively jumping spiders, along with the other five taxa of spiders observed, were found on one in five host plants and, therefore, represented an important potential threat to eggs and young instars. Spiders are important predators on monarch eggs and larvae (Smithers 1973; Zalucki and Kitching 1982; Lynch and Martin 1993; De Anda and Oberhauser 2015; Hermann et al. 2019; Myers 2019; Myers et al. 2020; Stevenson et al. 2021) and the stout growth form of *A. viridis* represents an attractive location for spiders to place webs and harborages (Vasconcellos-Neto et al. 2017). However, fire ants may be the most important predator of monarch eggs and larvae in the southern U.S. (Calvert 1996, 2004). On our study site the mound density of fire ants was 147 mounds per hectare, at the lower end of the average range of 155 to 470 mounds per hectare typical of North American populations (Macom and Porter 1996; Porter et al. 1997). Fire ants seemed to have variable effects on the survival of fall monarchs and queens and may have been most important during the very dry conditions of 2017. This could be due to the effect of weather conditions on food resources and the resulting changes in foraging strategies by the ants (Stein et al. 1990; Cassill and Tschinkel 1999; Vogt et al. 2002). However, even in 2017, survivorship was high enough that fire ants could not be attributed as having the catastrophic effects on monarch survival cited by other studies (Calvert 1996, 2004).

Queen butterflies in Texas are poorly studied. In our study, the occurrence of queens in fall corresponded well with that of monarchs, especially relative to peaks in September. Queens exhibit southward migratory movements in south Texas and Mexico in the fall (Einem 2003). However, northward movements of queens have not been studied. We found that for most of Texas east of the 100th meridian, there are two peaks in the abundance of queens. In south Texas, these peaks occur in January-February and again in October-November. In central and north Texas, these peaks occurred in July and October. Though we can't be certain, this pattern suggests an annual northward colonization of Texas followed by a southward migration in fall. Analyses of queens migrating southward through northern Mexico in October and November show that these migrants originate from north Texas and the southeastern U.S. (Hobson et al. 2021). Collectively, the phenology of queens across Texas suggest that queens engage in migratory movements that are similar, albeit on a much smaller scale, to that of monarchs. However, because queens are a mainly tropical and subtropical species, they may be poorly adapted to weather fluctuations at the northern periphery of their summer and fall distribution. This would explain why queens on our study site were far less abundant and had very low survivorship in the very dry fall of 2017 as compared to the much wetter fall of 2018. Variations in weather conditions and their effect on southern generations might also explain why queens frequently, but unpredictably, show up far to the north in the central plains (Lotts and Naberhaus 2021) and northeastern U.S. (Moskowitz 2003).

Monarchs and queens utilize the same host plants and can compete for access to these plants. In Florida, queens are displaced from ovipositing on preferred food plants by the presence of large numbers of monarchs (Brower 1961) and, reciprocally, monarchs are displaced from ovipositing on preferred food plants by the presence of large numbers of queens (Brower 1962). In our study it was not overtly evident that the two species were competing and, in both years, there were over 30 vacant host plants for every egg found. However, among 414 host plants that held eggs, 49 harbored multiple eggs; 44 with two eggs, four with three eggs, and one with four eggs. Multiple eggs on host plants in spite of the high

number of vacant plants implies the possibility that females are selective and that high-quality host plants might be limiting. Because they are a broad-ranging, highly mobile species, queens have a strong potential to expand into North America in response to climate change (Kocsis 2011). Furthermore, queens are less susceptible than are monarchs to the protozoan ectoparasite *Ophryocystis elektroscirrha* (OE) (Barriga et al. 2016) which gives them a strong competitive advantage. If climate change and parasite resistance favor queens, then fall reproduction by monarchs may be negatively impacted in the future.

The phenology of fall reproduction is important as it affects the survival of the resulting generation. At our latitude the average first frost occurs on November 11th, though freezing temperatures occasionally occur earlier as was observed in 2017. The peak in reproduction of monarchs and queens from September to mid-October in Texas is adaptive because it provides sufficient time for the great majority of the resulting adults to eclose before the advent of freezing weather (Baum and Sharber 2012). An earlier study found that most monarchs that eclose in fall in Texas are in diapause and show migratory behaviors (Batalden and Oberhauser 2015) and stable isotope data show that 11% to 30% of overwintering monarchs in Mexico originate from the southern great plains of the U.S. (Flockhart et al. 2017). Because of the high productivity identified in our study and the importance of fall reproduction toward overwintering recruitment, fall reproduction, and the health and productivity of the resulting offspring, must be an important component of the management of eastern monarch populations.

For monarchs, the temporal displacement between the peak in fall reproduction and the peak period of migrants observed in our study is consistent with other studies on fall breeding in Texas and Oklahoma (Prysby and Oberhauser 2004; Baum and Sharber 2012; Batalden and Oberhauser 2015) and supports the prevailing hypothesis that fall-breeding monarchs in the southern U.S. are early migrants that originated from adults that eclosed further north (Calvert 1999; Zalucki and Rochester 1999; Borland et al. 2004; Batalden and Oberhauser 2015; Satterfield et al. 2018; Majewska and Altizer 2019). However, in 2017, monarchs were observed in small numbers all summer in north Texas (JGK Pers. Obs.). In that year in particular, on our study site there were large numbers of reproductive monarchs in mid-August and the source of these individuals is uncertain. Winter temperatures at this latitude are too cold for monarchs to overwinter (Zalucki and Rochester 1999; Howard et al. 2010), so the summer monarchs might have hatched in the area in spring and failed to migrate or they could be individuals that diffused northward from the resident populations along the coast of the Gulf of Mexico, 400 km further south. Resident populations of monarch butterflies along the gulf coast of Texas appear to be a relatively recent phenomenon (Howard et al. 2010; Majewska and Altizer 2019) and the dynamics of these populations are largely unknown and need further study. It is possible that a few of our fall-breeding individuals were not migrants from further north.

In general, fall reproduction in monarchs is believed to be caused when migrants come out of diapause upon reaching the southern U.S. Among adults captured in fall in Minnesota and Wisconsin, only 3% of females had mated whereas among females captured in fall in Texas, over 17% had mated (Borland et al. 2004). The presence of viable populations of milkweed at these lower latitudes causes the adults to come out of diapause (Borland et al. 2004; Prysby and Oberhauser 2004; Baum and Sharber 2012). This tendency to come out of diapause may be exacerbated by the cultivation of tropical milkweed (*A. curassavica*) in gardens and landscaping plots. In Texas, fall egg densities were much higher in gardens planted with *A. curassavica* than in plots with native milkweeds (Prysby and Oberhauser 2004) and monarchs, when given a choice, prefer to oviposit on *A. curassavica* rather than on the native milkweeds *A. asperula* and *A. oenotheroides* (Batalden and Oberhauser 2015). In Georgia, females in fall were more likely to exhibit egg development when exposed to *A. curassavica* than when exposed to *A. incarnata* (Majewska and Altizer 2019). It has been proposed that the widespread cultivation of *A. curassavica* might be disrupting migratory patterns of monarchs (Malcolm 2018; Clement and Crawford 2020) and increasing their exposure to parasites (Satterfield et al. 2015; Malcolm 2018).

However, fall reproduction is also likely to be a normal part of the phenotypic plasticity of monarchs. Native milkweeds are widely available in late summer and fall in the southern U.S. (Tracy et al. 2021) and these plants are extensively utilized for

fall breeding by monarchs (Calvert 1999). Mowing (Baum and Mueller 2015; Knight et al. 2019; Dee and Baum 2019) and summer burns (Baum and Sharber 2012) cause *A. viridis* to sprout new growth. At our study site, mowing in late July generated new growth through September and into October, resulting in a milkweed density that was high compared to studies conducted in other parts of the monarch's distribution (Pleasants 2017; Kaul and Wilsey 2019). It is important to note that the studies conducted in Texas and Georgia on milkweed preferences cited in the preceding paragraph found that native milkweeds also stimulated fall reproduction. In the Texas study, male monarchs were as likely to exhibit mating behavior when caged with the native milkweed *A. oenotheroides* as they were when caged with *A. curassavica* (Batalden and Oberhauser 2015). In the Georgia study (Majewska and Altizer 2019), males exposed to *A. incarnata* were more likely to be reproductive than those exposed to *A. curassavica* and females exposed to *A. incarnata* were only slightly less likely to show reproductive behavior than were females exposed to *A. curassavica*. Majewska and Altizer (2019) suggested that a lower affinity of monarchs for *A. incarnata* was due to the low cardenolide concentrations found in that species as compared to *A. curassavica*. It would be useful to replicate the Georgia study using *A. viridis* because, unlike the other native species mentioned above, the cardenolide content and the cardenolide polarity profiles of *A. viridis* and *A. curassavica* are very similar (Rasmann and Agrawal 2011). Furthermore, *A. viridis* is abundant in Texas and Oklahoma in the fall (Baum and Sharber 2012; Dee and Baum 2019; Tracy et al. 2021) and at the landscape scale much more abundant than *A. curassavica* (Calvert 1999). Our study site contained only *A. viridis* and it seems unlikely that exposure to *A. curassavica* was an important factor in stimulating reproduction on our study site. It seems more plausible that fall reproduction in the presence of healthy populations of *A. viridis* is a normal adaptive response for monarchs (Freedman et al. 2018; Green 2021).

While the cultivation of exotic milkweeds should be discouraged, the importance of fall monarch reproduction on native milkweeds should not be ignored. Native milkweeds, most importantly *A. viridis*, are widely available in the southern plains in late summer and fall (Calvert 1999; Tracy et al. 2021). As such, fall reproduction is a normal aspect of the species' biology and likely represents an adaptive trade-off between the benefits of current reproduction and those of continuing migration to Mexico. Most importantly, fall reproduction buffers against reproductive failures in earlier generations (Flockhart et al. 2017). For these reasons, land management for successful fall breeding in the southern U.S. should be an important aspect of monarch recovery plans.

Implications For Conservation

Fall reproduction by monarchs is an important, yet neglected component for the management of the eastern monarch butterfly particularly given evidence that the offspring produced through fall reproduction contribute significantly to wintering populations in Mexico. This reproduction needs to be emphasized in monarch recovery plans. Land managers in the southern U.S. wanting to contribute to increased monarch recruitment need to coordinate prescribed burns and mowing schedules to maximize production of healthy *A. viridis* and other native milkweed plants in the fall. In Texas, mowing or burning should be conducted in July and should be avoided in September and October when monarch eggs and larvae occupy host plants.

Queen butterflies showed variable survival to the third instar in north Texas. However, the characteristics of this species make it likely that it will expand its distribution northward into the U.S. in response to climate change. Because queens could displace monarchs from favorable host plants, monitoring of this species and its populations is essential to understand the impact of the queen on future dynamics of fall reproduction in monarchs.

Declarations

Acknowledgements

We would like to thank Dr. Lani Lyman-Henley and Dr. Johanna Delgado-Acevedo for their time and advice on earlier versions of this study. Thanks, also to Roger Thigley and the Sulphur Springs Economic Development Corporation for use of the study site. Mike Quinn is thanked for assistance in arthropod identification. Lastly, we thank the Department of Biological and Environmental Sciences and the College of Science and Engineering at Texas A&M University – Commerce for financial support.

Funding

This project was funded by the Department of Biological and Environmental Sciences and by the College of Science and Engineering at Texas A&M University – Commerce. The APC was funded by Texas A&M University—Commerce. Additional funding came from the Texas Comptroller of Public Accounts, Economic Growth and Endangered Species Management Division, Contract Number 6192CS.

Author information

Alyx Scott, Houston Zoo, 6200 Herman Park Drive, Houston, Texas 77030, USA. Email: ascott@houstonzoo.org

Kelsey Contreras, Environmental Health and Safety, University of Texas at Arlington, Arlington, Texas 76019, USA. Email: Kelseyhelgerson7@gmail.com

Misty Stevenson, Dallas Arboretum and Botanical Garden, 8525 Garland Road, Dallas, Texas 75218, USA. Email: mnixon@dallasarboretum.org

Kalynn L. Hudman, Department of Biological and Environmental Sciences, Texas A&M University – Commerce, Commerce, Texas 75428, USA. Email: kalynnhudman@gmail.com

Jeffrey G. Kopachena, Department of Biological and Environmental Sciences, Texas A&M University – Commerce, Commerce, Texas 75428, USA. Email: Jeff.Kopachena@tamuc.edu

Ethics declarations

Conflicts of interest/Competing interests: The authors declare no conflicts of interest.

Availability of data and material: Data is available upon request from the corresponding author.

Code availability: Not applicable

Authors' contributions: All authors contributed to the study design, material preparation and data collection. Statistical analyses were performed by Alyx Scott and Jeffrey Kopachena. The first draft was composed by Alyx Scott and all authors commented on previous versions of the manuscript. All authors have read and have approved the submitted manuscript.

Ethics approval: The authors declare no financial or non-financial interests, have no competing interests to declare, and have no financial or proprietary interests in materials discussed in the manuscript. None of the authors have affiliations with, or involvement in, any organization or entity with any financial or nonfinancial interest in the subject matter or materials described in the manuscript.

Consent to participate: Not applicable

Consent for publication: Not applicable

References

1. Baker AM, Potter DA (2020) Invasive paper wasp turns urban pollinator gardens into ecological traps for monarch butterfly larvae. Sci Rep 10:1–7. https://doi.org/10.1038/s41598-020-66621-6
2. Barriga PA, Sternberg ED, Lefèvre T, et al (2016) Occurrence and host specificity of a neogregarine protozoan in four milkweed butterfly hosts (*Danaus* spp.). J Invertebr Pathol 140:75–82. https://doi.org/10.1016/j.jip.2016.09.003
3. Batalden RV, Oberhauser KS (2015) Potential changes in eastern North American monarch migration in response to an introduced milkweed, *Asclepias curassavica*. In: Monarchs in a changing world: Biology and conservation of an iconic insect. Cornell University Press, Ithaca, New York, pp 215–224.
4. Baum KA, Mueller EK (2015) Grassland and roadside management practices affect milkweed abundance and opportunities for monarch recruitment. In: Monarchs in a changing world: Biology and conservation of an iconic butterfly. Cornell University Press, Ithaca, NY, pp 197–202.
5. Baum KA, Sharber WV (2012) Fire creates host plant patches for monarch butterflies. Biol Lett 8:968–971. https://doi.org/10.1098/rsbl.2012.0550
6. Borkin SS (1982) Notes on Shifting Distribution Patterns and Survival of Immature *Danaus plexippus* (Lepidoptera: Danaidae) on the Food Plant Asclepias syriaca. Gt Lakes Entomol 15:199–206.
7. Borland J, Johnson C, Crumpton T, et al (2004) Characteristics of fall migratory monarch butterflies, *Danaus plexippus*, in Minnesota and Texas: Biology and Conservation. In: The Monarch Butterfly: Biology and Conservation. Cornell University Press, Ithaca, NY, pp 97–104.
8. Brower LP (1961) Studies on the migration of the monarch butterfly I. Breeding populations of *Danaus plexippus* and *D. gilippus berenice* in south central Florida. Ecology 42:76–83. https://doi.org/10.2307/1933269
9. Brower LP (1962) Evidence for interspecific competition in natural populations of the monarch and queen butterflies, *Danaus plexippus* and *D. gilippus berenice* in south central Florida. Ecology 43:549–552.
10. Brower LP, Williams EH, Dunford KS, et al (2018) A long-term survey of spring monarch butterflies in north-central Florida. J Nat Hist 52:2025–2046. https://doi.org/10.1080/00222933.2018.1510057
11. Brym MZ, Henry C, Kendall RJ (2018) Potential significance of fall breeding of the monarch butterfly (*Danaus plexippus*) in the rolling plains ecoregion of west Texas. Tex J Sci 70:1–7. https://doi.org/10.32011/txjsci_70_1_Note4
12. Brym MZ, Henry C, Lukashow-Moore SP, et al (2020) Prevalence of monarch (*Danaus plexippus*) and queen (*Danaus gilippus*) butterflies in West Texas during the fall of 2018. BMC Ecol 20:1–13. https://doi.org/10.1186/s12898-020-00301-x
13. Calvert WH (1996) Fire ant predation on monarch larvae (Nymphalidae: Danainae) in a central Texas prairie. J Lepidopterists Soc 50:149–151.
14. Calvert WH (1999) Patterns in the spatial and temporal use of Texas milkweeds (Asclepiadaceae) by the monarch butterfly (*Danaus plexippus* L.) during fall, 1996. J Lepidopterists Soc 53:37–44.
15. Calvert WH (2004) The effect of fire ants on monarchs breeding in Texas. In: The monarch butterfly: Biology and conservation. Cornell University Press, Ithaca, NY, pp 47–53.
16. Cassill DL, Tschinkel WR (1999) Regulation of Diet in the Fire Ant, *Solenopsis invicta*. J Insect Behav 12:307–328. https://doi.org/10.1023/A:1020835304713
17. Clement KA, Crawford PH (2020) Fall available tropical milkweed (*Asclepias curassavica* L.) may be a population sink for the monarch butterfly. Okla Native Plant Rec 20:68–78.
18. De Anda A, Oberhauser KS (2015) Invertebrate natural enemies and stage-specific mortality rates of monarch eggs and larvae. In: Monarchs in a changing world: biology and conservation of an iconic butterfly. Cornell University Press, Ithaca, NY, pp 60–70.
19. Dee JR, Baum KA (2019) Mowing Frequency Influences Number of Flowering Stems but not Population Age Structure of *Asclepias viridis*, an Important Monarch Host Plant. Am Midl Nat 182:27–35. https://doi.org/10.1674/0003-0031-182.1.27

20. Dockx C (2012) Differences in phenotypic traits and migratory strategies between eastern North American monarch butterflies, *Danaus plexippus* (L.). Biol J Linn Soc 106:717–736. https://doi.org/10.1111/j.1095-8312.2012.01916.x
21. Einem GE (2003) Fall Migration of Queens, *Danaus gilippus strigosus* Bates, in Mexico and Texas. News Lepidopterists Soc 45:39–47.
22. Fisher KE, Hellmich RL, Bradbury SP (2020) Estimates of common milkweed (*Asclepias syriaca*) utilization by monarch larvae (*Danaus plexippus*) and the significance of larval movement. J Insect Conserv 24:297–307. https://doi.org/10.1007/s10841-019-00213-2
23. Flockhart DTT, Brower LP, Ramirez MI, et al (2017) Regional climate on the breeding grounds predicts variation in the natal origin of monarch butterflies overwintering in Mexico over 38 years. Glob Change Biol 23:2565–2576. https://doi.org/10.1111/gcb.13589
24. Flockhart DTT, Pichancourt J-B, Norris DR, Martin TG (2015) Unravelling the annual cycle in a migratory animal: breeding-season habitat loss drives population declines of monarch butterflies. J Anim Ecol 84:155–165. https://doi.org/10.1111/1365-2656.12253
25. Flockhart DTT, Wassenaar LI, Martin TG, et al (2013) Tracking multi-generational colonization of the breeding grounds by monarch butterflies in eastern North America. Proc R Soc B Biol Sci 280:20131087. https://doi.org/10.1098/rspb.2013.1087
26. Freedman MG, Dingle H, Tabuloc CA, et al (2018) Non-migratory monarch butterflies, *Danaus plexippus* (L.), retain developmental plasticity and a navigational mechanism associated with migration. Biol J Linn Soc 123:265–278. https://doi.org/10.1093/biolinnean/blx148
27. Green DA (2021) Monarch Butterfly Migration as an Integrative Model of Complex Trait Evolution. Am Nat 198:142–157. https://doi.org/10.1086/714526
28. Greenland S, Mansournia MA, Altman DG (2016) Sparse data bias: a problem hiding in plain sight. BMJ Med 352:1–6. http://dx.doi.org/10.1136/bmj.i1981
29. Grodsky SM, Saul-Gershenz LS, Moore-O'Leary KA, Hernandez RR (2020) Her Majesty's Desert Throne: The Ecology of Queen Butterfly Oviposition on Mojave Milkweed Host Plants. Insects 11:257. https://doi.org/10.3390/insects11040257
30. Gustafsson KM, Agrawal AA, Lewenstein BV, Wolf SA (2015) The Monarch Butterfly through Time and Space: The Social Construction of an Icon. BioScience 65:612–622. https://doi.org/10.1093/biosci/biv045
31. Haan NL, Landis DA (2019) Grassland disturbance increases monarch butterfly oviposition and decreases arthropod predator abundance. Biol Conserv 233:185–192. https://doi.org/10.1016/j.biocon.2019.03.007
32. Hermann SL, Blackledge C, Haan NL, et al (2019) Predators of monarch butterfly eggs and neonate larvae are more diverse than previously recognised. Sci Rep 9:1–9. https://doi.org/10.1038/s41598-019-50737-5
33. Hobson K, Kusack J, Mora-Alvarez B (2021) Origins of Six Species of Butterflies Migrating through Northeastern Mexico: New Insights from Stable Isotope (δ2H) Analyses and a Call for Documenting Butterfly Migrations. Diversity 13:102. https://doi.org/10.3390/d13030102
34. Howard E, Aschen H, Davis AK (2010) Citizen Science Observations of Monarch Butterfly Overwintering in the Southern United States. Psyche (Stuttg) 2010:e689301. https://doi.org/10.1155/2010/689301
35. Hudman KL (2018) An Experimental Study of the Direct and Indirect Effects of Red Imported Fire Ants (*Solenopsis invicta*) on Monarch Butterfly (*Danaus plexippus*) Eggs and Larvae and the Arthropod Community on Milkweed Plants (*Asclepias viridis*) - ProQuest. https://www.proquest.com/openview/4b557182976e5243bf6e81e8caed156a/1?pq-origsite=gscholar&cbl=18750&diss=y. Accessed 1 Jun 2022
36. Kaul AD, Wilsey BJ (2019) Monarch butterfly host plant (milkweed *Asclepias* spp.) abundance varies by habitat type across 98 prairies. Restor Ecol 27:1274–1281. https://doi.org/10.1111/rec.12993

37. Knight A, Brower LP (2009) The Influence of Eastern North American Autumnal Migrant Monarch Butterflies (*Danaus plexippus* L.) on Continuously Breeding Resident Monarch Populations in Southern Florida. J Chem Ecol 35:816–823. https://doi.org/10.1007/s10886-009-9655-z
38. Knight SM, Norris DR, Derbyshire R, Flockhart DTT (2019) Strategic mowing of roadside milkweeds increases monarch butterfly oviposition. Glob Ecol Conserv 19:e00678. https://doi.org/10.1016/j.gecco.2019.e00678
39. Koch RL, Venette RC, Hutchison WD (2005) Influence of Alternate Prey on Predation of Monarch Butterfly (Lepidoptera: Nymphalidae) Larvae by the Multicolored Asian Lady Beetle (Coleoptera: Coccinellidae). Environ Entomol 34:410–416. https://doi.org/10.1603/0046-225X-34.2.410
40. Kocsis M, Hufnagel L (2011) Impacts of climate change on lepidoptera species and communities. Appl Ecol Environ Res 9:43–72. https://doi.org/10.15666/aeer/0901_043072
41. Lotts K, Naberhaus T (2021) *Danaus gilippus*. In: Butterflies Moths N. Am. http://www.butterfliesandmoths.org/
42. Lynch SP, Martin RA (1993) Milkweed host plant utilization and cardenolide sequestration by monarch butterflies in Louisiana and Texas. In: Biology and conservation of the monarch butterfly. Natural History Museum of Los Angeles County, Los Angeles, California, USA, pp 107–123.
43. Macom TE, Porter SD (1996) Comparison of polygyne and monogyne red imported fire ant (Hymenoptera: Formicidae) population densities. Ann Entomol Soc Am 89:535–543. https://doi.org/10.1093/aesa/89.4.535
44. Majewska AA, Altizer S (2019) Exposure to Non-Native Tropical Milkweed Promotes Reproductive Development in Migratory Monarch Butterflies. Insects 10:253. https://doi.org/10.3390/insects10080253
45. Malcolm SB (2018) Anthropogenic Impacts on Mortality and Population Viability of the Monarch Butterfly. Annu Rev Entomol 63:277–302. https://doi.org/10.1146/annurev-ento-020117-043241
46. Malcolm SB, Cockrell BJ, Brower LP (1993) Spring recolonization of eastern North America by the monarch butterfly: successive brood or single sweep migration. In: Biology and conservation of the monarch butterfly. Natural History Museum of Los Angeles County, Los Angeles, California, pp 253–267.
47. McCoshum SM, Andreoli SL, Stenoien CM, et al (2016) Species distribution models for natural enemies of monarch butterfly (*Danaus plexippus*) larvae and pupae: distribution patterns and implications for conservation. J Insect Conserv 20:223–237. https://doi.org/10.1007/s10841-016-9856-z
48. McLaughlin RE, Myers J (1970) *Ophryocystis elektroscirrha* sp. n., a Neogregarine Pathogen of the Monarch Butterfly *Danaus plexippus* (L.) and the Florida Queen Butterfly *D. gilippus berenice* Cramer. J Protozool 17:300–305. https://doi.org/10.1111/j.1550-7408.1970.tb02375.x
49. Monarch Joint Venture (2021) 2021 Monarch Conservation Implementation Plan. https://monarchjointventure.org/images/uploads/documents/2021_Monarch_Conservation_Implementation_Plan.pdf
50. Moranz R, Brower LP (1998) Geographic and temporal variation of cardenolide-based chemical defenses of queen butterfly (*Danaus gilippus*) in northern Florida. J Chem Ecol 24:905–932. https://doi.org/10.1023/A:1022329702632
51. Moskowitz D P (2003) The Queen (*Danaus gilippus* Cramer) dilemma in the Northeastern United States. News Lepidopterists Soc 45:62–36.
52. Mueller EK, Baum KA (2014) Monarch–parasite interactions in managed and roadside prairies. J Insect Conserv 18:847–853. https://doi.org/10.1007/s10841-014-9691-z
53. Myers AT (2019) The interacting influences of habitat context and predators on monarch butterfly (Danaus plexippus L.) Oviposition and survival in agricultural landscapes. Doctoral Dissertation, Michigan State University
54. Myers A, Bahlai CA, Landis DA (2019) Habitat Type Influences *Danaus plexippus* (Lepidoptera: Nymphalidae) Oviposition and Egg Survival on *Asclepias syriaca* (Gentianales: Apocynaceae). Environ Entomol 48:675–684. https://doi.org/10.1093/ee/nvz046
55. Myers AT, Haan NL, Landis DA (2020) Video surveillance reveals a community of largely nocturnal *Danaus plexippus* (L.) egg predators. J Insect Conserv 24:731–737. https://doi.org/10.1007/s10841-020-00248-w

56. Nail K (2020) Monarch (*Danaus plexippus*) Species Status Assessment Report, version 2.1 September 2020. 126
57. Nestle R, Daniels JC, Dale AG (2020) Mixed-Species Gardens Increase Monarch Oviposition without Increasing Top-Down Predation. Insects 11:648. https://doi.org/10.3390/insects11090648
58. Oberhauser K, Wiederholt R, Diffendorfer JE, et al (2017) A trans-national monarch butterfly population model and implications for regional conservation priorities: Regional monarch conservation priorities. Ecol Entomol 42:51–60. https://doi.org/10.1111/een.12351
59. Oberhauser KS, Anderson M, Anderson S, et al (2015) Lacewings, wasps, and flies—oh my: insect enemies take a bite out of monarchs. Monarchs Chang World Biol Conserv Iconic Insect Cornell Univ Press Ithaca NY 71–82.
60. Oberhauser KS, Prysby MD, Mattila HR, et al (2001) Temporal and spatial overlap between monarch larvae and corn pollen. Proc Natl Acad Sci 98:11913–11918. https://doi.org/10.1073/pnas.211234298
61. Pleasants J (2017) Milkweed restoration in the Midwest for monarch butterfly recovery: estimates of milkweeds lost, milkweeds remaining and milkweeds that must be added to increase the monarch population. Insect Conserv Divers 10:42–53. https://doi.org/10.1111/icad.12198
62. Porter SD, Williams DF, Patterson RS, Fowler HG (1997) Intercontinental differences in the abundance of *Solenopsis* fire ants (Hymenoptera: Formicidae): escape from natural enemies? Environ Entomol 26:373–384. https://doi.org/10.1093/ee/26.2.373
63. Prysby MD (2004) Natural enemies and survival of monarch eggs and larvae. In: The monarch butterfly: biology and conservation. Cornell University Press, Ithaca, NY, pp 27–37.
64. Prysby MD, Oberhauser KS (2004) Temporal and Geographic Variation in Monarch Densities: Citizen Scientists Document Monarch Population Patterns. In: The Monarch Butterfly: Biology and Conservation. Cornell University Press, Ithaca, NY, pp 9–20.
65. Rasmann S, Agrawal AA (2011) Latitudinal patterns in plant defense: evolution of cardenolides, their toxicity and induction following herbivory: Latitude and the evolution of plant defense. Ecol Lett 14:476–483. https://doi.org/10.1111/j.1461-0248.2011.01609.x
66. Rawlins JE, Lederhouse RC (1981) Developmental Influences of Thermal Behavior on Monarch Caterpillars (*Danaus plexippus*): An Adaptation for Migration (Lepidoptera: Nymphalidae: Danainae). J Kans Entomol Soc 54:387–408.
67. Ritland DB (1991a) Unpalatability of Viceroy butterflies (*Limenitis archippus*) and their purported mimicry models, Florida Queens (*Danaus gilippus*). Oecologia 88:102–108. https://doi.org/10.1007/BF00328409
68. Ritland DB (1991b) Revising a classic butterfly mimicry scenario: demonstration of Müllerian mimicry between Florida viceroys (*Limenitis archippus floridensis*) and queens (*Danaus gilippus berenice*). Evolution 45:918–934. https://doi.org/10.1111/j.1558-5646.1991.tb04360.x
69. Sakai WH (1993) Notes on *Danaus gilippus strigosus* (Nymphalidae: Danainae) in southern California. J Lepidopterists' Soc 47:160–161.
70. Satterfield DA, Maerz JC, Altizer S (2015) Loss of migratory behaviour increases infection risk for a butterfly host. Proc R Soc B Biol Sci 282:20141734. https://doi.org/10.1098/rspb.2014.1734
71. Satterfield DA, Maerz JC, Hunter MD, et al (2018) Migratory monarchs that encounter resident monarchs show life-history differences and higher rates of parasite infection. Ecol Lett 21:1670–1680. https://doi.org/10.1111/ele.13144
72. Saul-Gershenz L, Grodsky SM, Hernandez RR (2020) Ecology of the Western Queen Butterfly *Danaus gilippus thersippus* (Lepidoptera: Nymphalidae) in the Mojave and Sonoran Deserts. Insects 11:315. https://doi.org/10.3390/insects11050315
73. Scott A (2019) Survival of Fall Monarch (*Danaus plexippus*) and Queen (*Danaus gilippus*) Butterfly Eggs and Larvae in North Texas. Masters Thesis, Texas A&M University - Commerce
74. Semmens BX, Semmens DJ, Thogmartin WE, et al (2016) Quasi-extinction risk and population targets for the Eastern, migratory population of monarch butterflies (*Danaus plexippus*). Sci Rep 6:23265. https://doi.org/10.1038/srep23265

75. Sheehan N, Weber-Grullon L (2021) Journey North - Monarch Butterfly and Milkweed observations by volunteer community scientists across Central and North America (1996-2020) ver 1. In: Journey North - Monarch Butterfly Milkweed Obs. Volunt. Community Sci. Cent. N. Am. 1996-2020 Ver 1 Environ. Data Initiat. https://doi.org/10.6073/pasta/f7d7bef57f94b33b8a18a26954252412. Accessed 30 Sep 2021
76. Smithers CN (1973) A note on natural enemies of "*Danaus plexippus*" (L.) (Lepidoptera: Nymphalide) in Australia. Aust Entomol 1:37–40. https://search.informit.org/doi/10.3316/informit.141596079006793
77. Stein MB, Thorvilson HG, Johnson JW (1990) Seasonal changes in bait preference by red imported fire ant, *Solenopsis invicta* (Hymenoptera: Formicidae). Fla Entomol 73:117–123. https://doi.org/10.2307/3495334
78. Stevenson M, Hudman KL, Scott A, et al (2021) High Survivorship of First-Generation Monarch Butterfly Eggs to Third Instar Associated with a Diverse Arthropod Community. Insects 12:567. https://doi.org/10.3390/insects12060567
79. Thogmartin WE, Wiederholt R, Oberhauser K, et al (2017) Monarch butterfly population decline in North America: identifying the threatening processes. R Soc Open Sci 4:170760. https://doi.org/10.1098/rsos.170760
80. Tracy JL, Kantola T, Baum KA, Coulson RN (2021) Distribution, phenology, and land cover preference risk assessments of four South-Central US milkweeds (*Asclepias* spp.) important for monarch butterflies. Res Sq. https://doi.org/10.21203/rs.3.rs-755161/v1
81. Vasconcellos-Neto J, Messas YF, da Silva Souza H, et al (2017) Spider–plant interactions: an ecological approach. In: Behaviour and ecology of spiders. Springer, pp 165–214.
82. Vidal O, Rendón-Salinas E (2014) Dynamics and trends of overwintering colonies of the monarch butterfly in Mexico. Biol Conserv 180:165–175. https://doi.org/10.1016/j.biocon.2014.09.041
83. Vogt JT, Grantham RA, Corbett E, et al (2002) Dietary Habits of *Solenopsis invicta* (Hymenoptera: Formicidae) in Four Oklahoma Habitats. Environ Entomol 31:47–53. https://doi.org/10.1603/0046-225X-31.1.47
84. Voorhies KJ, Szymanski J, Nail KR, Fidino M (2019) A Method to Project Future Impacts From Threats and Conservation on the Probability of Extinction for North American Migratory Monarch (*Danaus plexippus*) Populations. Front Ecol Evol 7:. https://doi.org/10.3389/fevo.2019.00384
85. York HA, Oberhauser KS (2002) Effects of Duration and Timing of Heat Stress on Monarch Butterfly (*Danaus plexippus*) (Lepidoptera: Nymphalidae) Development. J Kans Entomol Soc 75:290–298.
86. Zalucki MP, Brower LP, Malcolm SB (1990) Oviposition by *Danaus plexippus* in relation to cardenolide content of three *Asclepias* species in the southeastern USA. Ecol Entomol 15:231–240. https://doi.org/10.1111/j.1365-2311.1990.tb00804.x
87. Zalucki MP, Clarke AR, Malcolm SB (2002) Ecology and Behavior of First Instar Larval Lepidoptera. Annu Rev Entomol 47:361–393. https://doi.org/10.1146/annurev.ento.47.091201.145220
88. Zalucki MP, Kitching RL (1982) Temporal and spatial variation of mortality in field populations of *Danaus plexippus* L. and *D. chrysippus* L. Larvae (Lepidoptera: Nymphalidae). Oecologia 53:201–207. https://doi.org/10.1007/BF00545664
89. Zalucki MP, Malcolm SB, Paine TD, et al (2001) It's the first bites that count: Survival of first-instar monarchs on milkweeds. Austral Ecol 26:547–555
90. Zalucki MP, Rochester WA (1999) Estimating the effect of climate on the distribution and abundance of *Danaus plexippus*: a tale of two continents. In: 1997 North American Conference on the Monarch Butterfly. Commission for Environmental Cooperation, Morelia, Michoacán, Mexico, pp 151–163
91. Zanden HBV, Chaffee CL, González-Rodríguez A, et al (2018) Alternate migration strategies of eastern monarch butterflies revealed by stable isotopes. Anim Migr 5:74–83. https://doi.org/10.1515/ami-2018-0006
92. Zipkin EF, Ries L, Reeves R, et al (2012) Tracking climate impacts on the migratory monarch butterfly. Glob Change Biol 18:3039–3049. https://doi.org/10.1111/j.1365-2486.2012.02751.x

93. Zylstra ER, Ries L, Neupane N, et al (2021) Changes in climate drive recent monarch butterfly dynamics. Nat Ecol Evol 5:1–12. https://doi.org/10.1038/s41559-021-01504-1

Tables

Table 1. Weather data for the study site compared with long-term normals. Weather data was from a NOAA weather station 2.3 km north of the study site. Long-term normal was based on data collected at this station during the 30-year period from 1981 to 2010.

	Precipitation (cm)[a]			Average Maximum Temperature (ºC)			Average Minimum Temperature (ºC)		
	2017	2018	Normal	2017	2018	Normal	2017	2018	Normal
August	30.30	3.61	6.20	32.26	37.32	33.94	23.21	25.09	22.00
September	0.03	20.24	7.59	32.01	30.61	30.06	18.51	21.59	17.61
October	9.45	19.61	13.69	26.79	23.85	24.06	11.61	14.76	11.50

[a] Precipitation expressed as total for each month, normal refers to average monthly totals.

Table 2. Stepwise logistic regression analysis of the effects of arthropod groups on the survival of fall monarch and queen eggs or larvae. Stepwise selection was used to generate the best subset of models using a p-value of 0.50 for entry or removal of a parameter from the model. Models are sorted in order of ascending AICc. Best model is based on minimum corrected AIC score (AICc), w_i is the Akaike weight of each model.

Model	AICc	$_{\Delta}$AICc	w_i	Likelihood Ratio X^2	Model Probability
Fire Ants, Jumping Spiders, All Other Non-Predatory Arthropods, Oleander Aphids	137.621	0.000	0.308	10.4318	0.0337
Fire Ants, Jumping Spiders, All Other Non-Predatory Arthropods	138.299	0.678	0.219	7.5477	0.0563
Fire Ants	138.540	0.919	0.194	3.0245	0.0820
Fire Ants, Jumping Spiders	138.962	1.341	0.157	4.7223	0.0943
Intercept Only	139.485	1.864	0.121	-	-
Summary of the best fit model. Concordance of this model was 46.3%.					
Parameter	DF	Maximum Likelihood Estimate	Standard Error	Wald Chi-Square	p-value
Intercept	1	-0.4180	0.2331	3.2162	0.0729
Fire Ants	1	-0.4913	0.4464	1.2110	0.2711
Jumping Spiders	1	-1.0568	0.7042	2.2523	0.1334
All Other Non-Predatory Arthropods	1	0.1185	0.0741	2.5550	0.1099
Oleander Aphids	1	-0.0284	0.0374	0.5765	0.4477

Table 3. Egg and larval survival relative to fire ant mound densities. Numbers represent the mean ± standard error of the number of fire ant mounds within 4m of host plants. Numbers in parentheses are sample sizes.

	Number of Mounds Within 4m of Host Plant		Kruskal – Wallis Test (C^2 Approximation)	
	Survived	Died	C^2	p
All Eggs – 2017	0.17 ± 0.11 (12)	0.74 ± 0.09 (219)	2.8765	0.0899
All Eggs – 2018	0.80 ± 0.14 (41)	0.77 ± 0.08 (190)	0.3024	0.5824
All Eggs – Both Years	0.66 ± 0.12 (53)	0.75 ± 0.06 (409)	0.0003	0.9873
Monarch Larvae[a]	0.35 ± 0.12 (26)	0.62 ± 0.08 (105)	2.3614	0.1244
Queen Larvae[a]	0.96 ± 0.19 (27)	0.86 ± 0.16 (51)	0.5348	0.4646

[a] Includes only those individuals hatched from focal eggs that could be identified to species at the first instar. Data are combined for both years of the study.

Figures

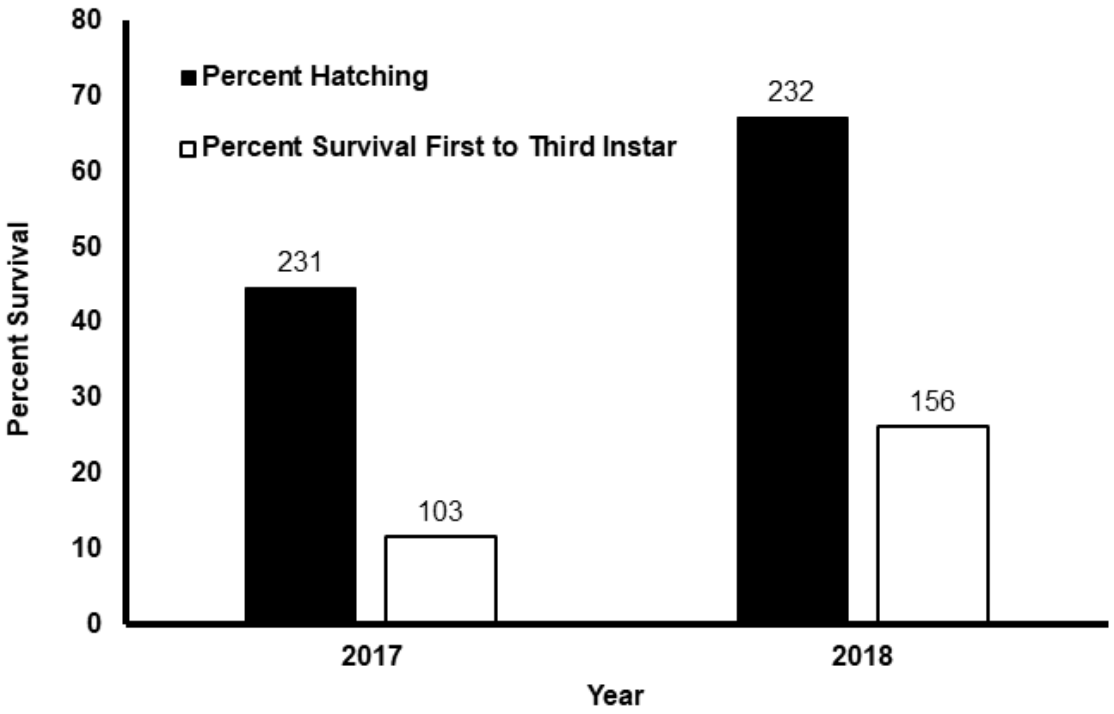

Figure 1

Percent of all eggs that hatched or survived from first to third instar. These data combine monarchs and queens. In 2017, 44.6% of eggs hatched. In 2018, 67.2% of eggs hatched, 51% more than had hatched in 2017 (Chi-square 2x2 Contingency Table; X^2 = 16.25, df = 1, p < 0.0001). Survival from hatching to third instar was 11.7% in 2017. Survival from hatching to

third instar in 2018 was 26.3%, 125% higher than it was in 2017 (Chi-square 2x2 Contingency Table; X^2 = 16.13, df = 1, p < 0.0001). Numbers over bars indicate the total number of eggs or first instars observed in each year

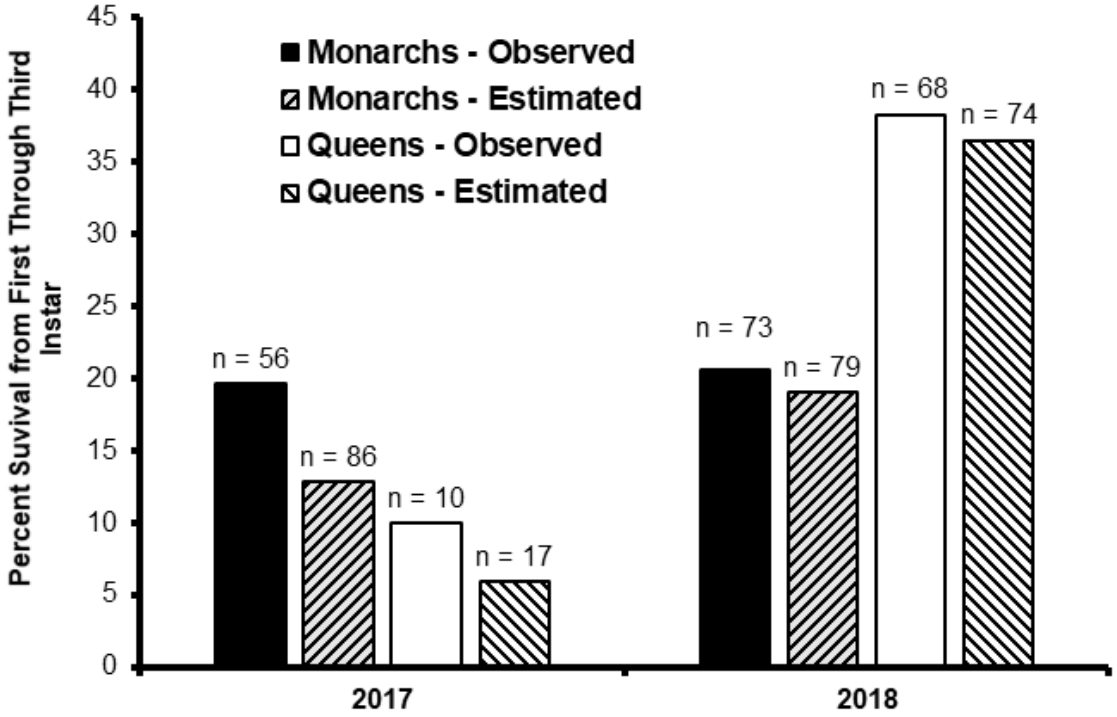

Figure 2

Percent survival from first instar to third instar by species. Among first instars identified to species, monarch survival was 19.6% in 2017 and 20.5% in 2018 (2 x 2 Contingency Table, X^2 = 0.161, df = 1, p = 0.8989). The survivorship of queens, varied from 10% in 2017 to 38.2% in 2018 (2 x 2 Contingency Table, Fisher's Exact Test, p = 0.1514). However, when survival was estimated by including larvae that died prior to identification (see text), monarch survivorship from hatching to third instar was 12.8% in 2017 and 19% in 2018 (2 x 2 Contingency Table, X^2 = 0.1911, df = 1, p = 0.2751). The estimated survival of queens was 5.9% in 2017 and 36.5% in 2018 (2 x 2 Contingency Table, X^2 = 6.0783, df = 1, p = 0.0137)

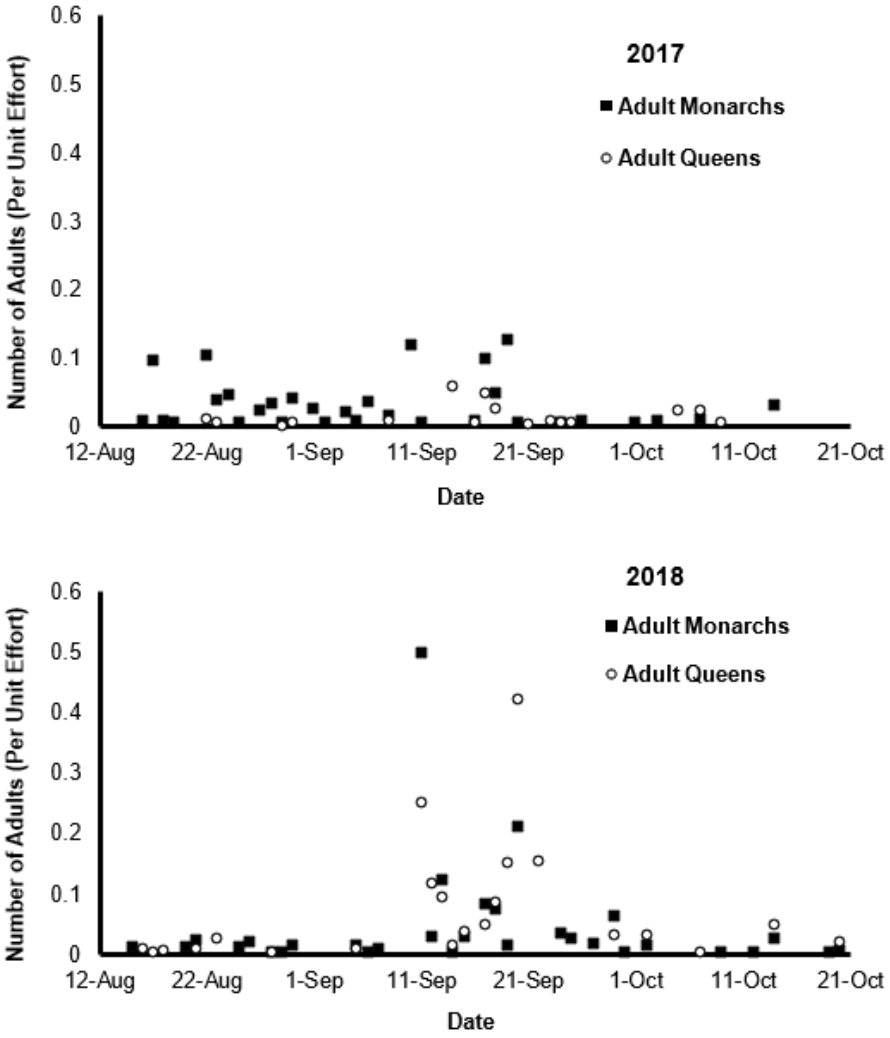

Figure 3

Study site occupancy by adult monarchs and queens during 2017 (upper graph) and 2018 (lower graph). The scales on the vertical axes are fixed to emphasize differences in abundance between years. In 2017, the average date of occupancy by monarchs was 12 days earlier than that of queens (t-test, Satterthwaite's method for unequal variances; $t = 3.06$, $df = 35.8$, $p = 0.0042$). In 2018, the average date of site occupancy by monarchs was three days earlier than that of queens (t-test, Pooled variance estimate; $t = 1.12$, $df = 112$, $p = 0.2692$). In 2017, monarchs were more abundant than queens; 80% of the adults were monarchs (t-test, Satterthwaite's method for unequal variances; $t = 3.04$, $df = 68.9$, $p = 0.0033$). In 2018, across the entire season, there were approximately equal numbers of monarchs and queens (t-test, Pooled variance estimate; $t = 0.25$, $df = 112$, $p = 0.8067$)

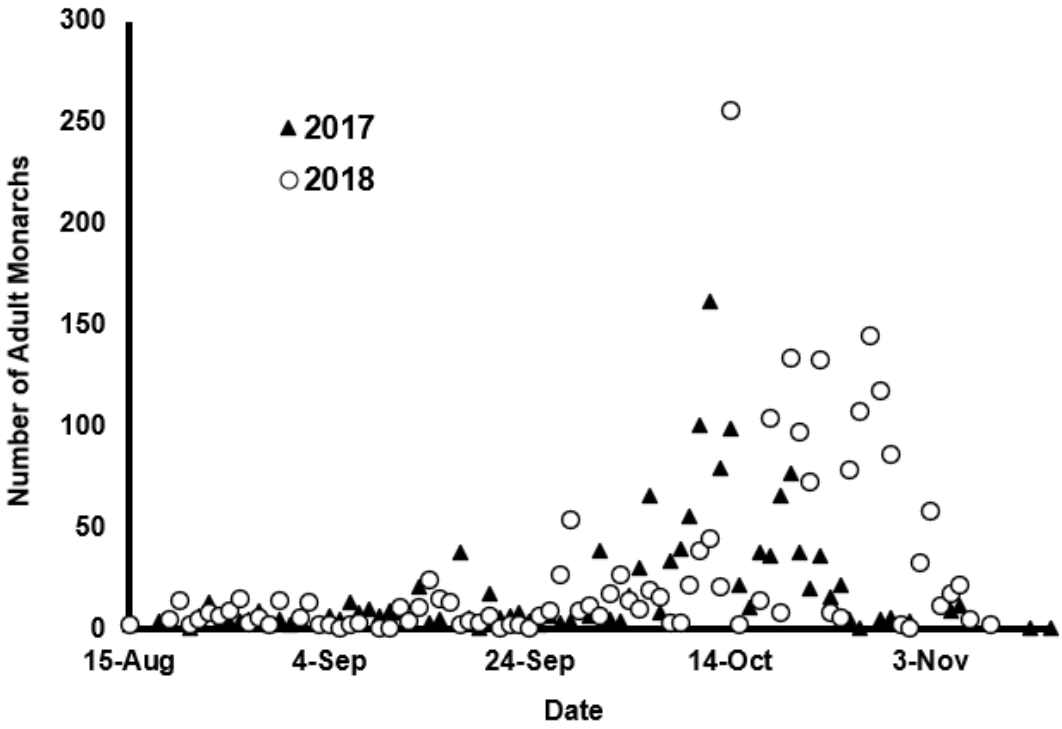

Figure 4

Journey North Data on monarch observations in the region around the study area from 15 August 2017 through 15 November 2017 and from 15 August 2018 through 15 November 2018. Data include only sightings of adults and do not include roost data. Solid triangles (n = 76 days) represent observations made in 2017 whereas open circles (n = 80 days) represent observations made in 2018. The average date for monarchs passing through the region was 7 October in 2017 and 15 October in 2018

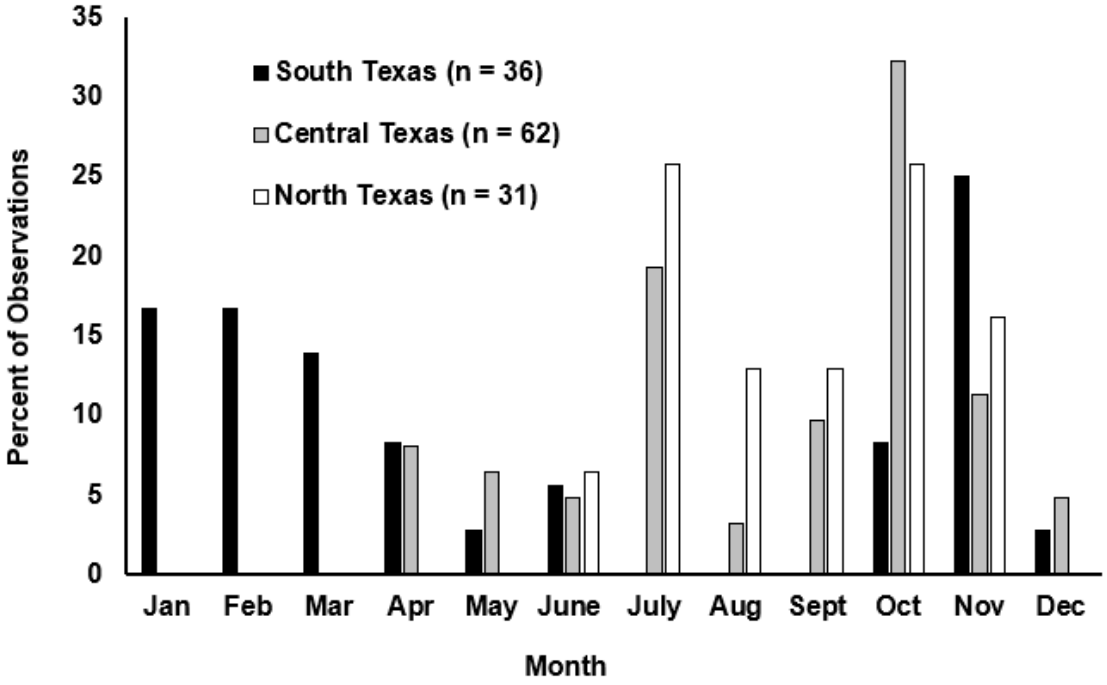

Figure 5

The phenology of queen butterflies among geographic regions in Texas based on data provided by Butterflies and Moths of North America (BAMONA). Data include 129 sightings of adults between 1 January 2000 and 31 December 2020. Geographic regions are defined in the methods section of the text. The pattern is consistent with a seasonal colonization of Texas starting in south Texas in winter and early spring and moving northward in two successive generations that peak in July and October in central and north Texas

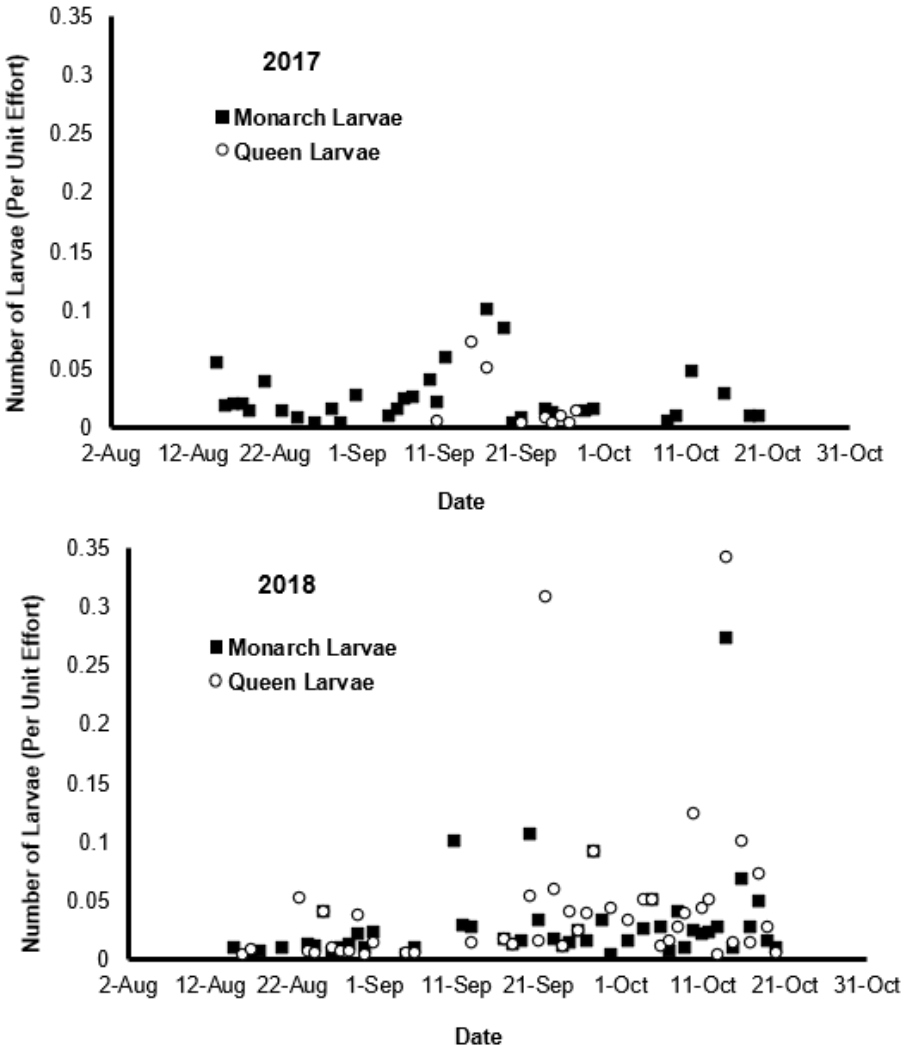

Figure 6

Study site occupancy by non-focal monarch and queen larvae in 2017 (upper graph) and 2018 (lower graph). The scales on the vertical axes are fixed to emphasize differences in abundance between years. In 2017, the average date for monarch larvae was six days earlier than it was for queen larvae (t-test, Satterthwaite's method for unequal variances; t = 1.79, df = 39.9, p = 0.0816). In 2018, the average date for monarch larvae was only two days earlier than it was for queen larvae (t-test, Pooled variance estimate; t = 0.65, df = 92, p = 0.5191). As observed for adults, in 2017, monarch larvae were much more abundant than queen larvae; 82% of the larvae were monarchs (t-test, Satterthwaite's method for unequal variances; t = 3.40, df = 86.13, p = 0.0010). However, in 2018, across the entire season, there were approximately equal numbers of monarch larvae and queen larvae (t-test, Satterthwaite's method for unequal variances; t = 0.88, df = 96.42, p = 0.3807)

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Additionalinformation.docx