UNIVERSITY OF CALIFORNIA, SANTA CRUZ

CAUSES OF MORTALITY TO AN OVERWINTERING POPULATION OF WESTERN MONARCH BUTTERFLIES (*Danaus plexippus*) AT LIGHTHOUSE FIELD IN SANTA CRUZ, CALIFORNIA

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ABSTRACT:

Monarch butterfly (*Danaus plexippus*) populations have decreased dramatically over the past two decades. Their populations are threatened by habitat loss, pesticide use, climate change, and disease. I worked with Groundswell Coastal Ecology to conduct a mortality study at Lighthouse Field State Beach (LHFSB) in Santa Cruz, California to calculate a minimum mortality rate for the 2017-2018 overwintering population. We collected 1,711 dead monarchs (755 females and 956 males) in the overwintering grove between November 2017 and March 2018. We estimated a minimum mortality of 12.6% for the 2017-2018 overwintering season using a population count of 13,533 (Xerces Society January Count Data). We assessed the probable cause of mortality for all whole dead monarchs collected (n=864). Our findings suggest that yellowjackets (*Vespula spp*) (49.2%) and avian predators (11.0%) were the most common causes of mortality for whole dead monarchs. We were unable to identify the source of mortality for the remaining 39.2% of whole dead individuals. Both males and females displayed similar temporal trends of mortality over the course of the overwintering season. We recommend that land managers control yellowjackets hives near monarch populations and continue mortality assessments at LHFSB to evaluate long-term efficacy of mortality reduction actions. This study will help inform land managers on effective restoration and conservation practices to decrease overwintering mortality rates for the monarch butterfly.

KEYWORDS: Monarch butterfly, *Danaus plexippus*, Santa Cruz, overwintering, mortality, threats, migration, conservation, management, yellowjacket, *Vespula spp*.

Introduction

In North America monarch butterflies (*Danaus plexippus plexippus*) are comprised of distinct Eastern and Western populations. The Eastern population overwinters in the mountains of Mexico, and the Western population overwinters in coastal California and southern Oregon (Brower and Malcom 1991). Monarch populations have experienced heavy declines since 2000 and are proposed for listing under the Endangered Species Act. Primary threats include habitat loss, pesticide use, climate change, and disease (Watanabe 2014). In California, monarchs depend heavily on migratory pathways and the forested groves of the Pacific coast that serve as critical overwintering habitat. Overwintering populations have declined by 50% since 1997 in the western monarch population (Jepsen and Black 2015), which has prompted efforts to improve overwintering survival by restoring monarch overwintering habitat (Schultz et al. 2017). Specifically, microhabitat conditions that provide high humidity, shade, freshwater, nectar sources, and protection from freezing temperatures and high winds are vital to promoting overwintering survival of monarchs (Jepsen et al. 2015).

Several factors are thought to have led to the decline of monarchs. One factor is the presence and availability of their host plant, milkweed (*Asclepias* spp.). Milkweed plants serve a vital role in the non-overwintering reproductive stages of monarchs. It is believed that milkweed loss has contributed largely to the decline of monarch populations (Oberhauser et al. 2015). Once the overwintering months pass and spring arrives in California, many monarchs fly inland in search of milkweed. Many sites in the Central Valley of California where milkweed was historically present have experienced substantial urban, suburban and agricultural development threatening monarch abundance and presence (Borders and Lee-Mäder 2015).

The protozoan parasite, *Ophryocystis elektroscirrha* (OE), further threatens monarchs. Monarchs are infected with OE through direct transfer from other monarchs and from OE on host milkweed plants. OE is transferred to offspring at birth from the infected female who lays eggs that contain OE spores. Once the caterpillar hatches from the egg, the new caterpillar digests OE spores. The OE spores are dormant until they reach the digestive tract of the caterpillar, where digestive chemicals break dormancy after which the parasites reproduce asexually. During the pupal stage, OE continues to reproduce and eventually forms spores. Once the newly emerged butterfly emerges from the chrysalis, it has dormant OE spores over its body, where the cycle begins again (Altizer et al. 2004). OE compromises the emerging adult and infected monarchs are too weak to fully expand their wings and hold on to their chrysalis. They often then fall to the ground and eventually die. It has not been proven that the infection of OE affects a female monarch's ability to reproduce, however some males that are infected with OE have difficulty mating because they are weak in comparison to unaffected males (Altizer and Oberhauser 1999).

The cardenolides that occur in milkweed are sequestered by the monarch and stored in the wings, as well as the rest of the body, deterring predators from preying on these insects (Anurag 2012). However, Brower (1988) studied an overwintering population in Mexico and concluded that two species of birds, the Baltimore Orioke (*Icterus galbula abeillei*) and the black-headed grosbeak (*Pheucticus melanocephalus*), consumed an estimated two million butterflies during the overwintering season because of their ability to digest large amounts of cardenolides. Other avian predators also threaten monarch butterflies, individuals having low levels of cardenolides may be of increased risk to predation.

Rodents are also known predators of monarch butterflies (Henry et al. 2017). Rodents prey on grounded monarchs, consuming their body and leaving behind the wings, which have little nutritional value and contain the largest concentration of cardenolides. Wasps are generalist predators that feed on monarch butterflies and other herbivorous insects (Leong et al 1990). Leong et al. (1990) studied at wasp predation on monarchs and reported that these predators have a large impact on monarchs. In 2017, a newspaper article from California reported adult monarchs that displayed signs of predation from unknown predators (Kipling 2017). In the article, Art Shapiro, professor of biology and ecology at UC Davis, stated that wasps attack monarchs in a similar manner.

Lighthouse Field State Beach in Santa Cruz, California was among the four largest Western populations in 2017 and is currently ranked the 7th most important site for monarch conservation and restoration out of 111 overwintering sites in California (Pelton et al. 2016). Since data collection began in 1997 at Lighthouse Field State Beach, monarch populations have fluctuated annually ranging from a minimum of 2,607 in 2008 and a maximum of 70,000 in 1997, with overall declines of 82% (Western Monarch Count Resource Center, 2018, Figure 1).

Groundswell Coastal Ecology, a Santa Cruz based non-profit organization, the Xerces Society for Invertebrate Conservation and the U.S. Fish and Wildlife Service Coastal Program developed a management plan to enhance grove attributes and overwintering monarch survival (Pelton 2017). The plan concludes that monarch conservation requires a variety of approaches, including strategic tree planting, increasing availability of native nectar sources, as well as promoting public engagement. The plan also identified the need for more information on the predators and development of a predator mitigation plan. We monitored overwintering monarch mortality and sources to better understand threats to the Lighthouse Field overwintering population.

Materials and Methods

I worked with members of Groundswell Coastal Ecology to monitor the overwintering migration of monarchs at Lighthouse Field State Beach in Santa Cruz from November 2017 through March 2018. The overwintering grove consists mainly of Monterey cypress tress (*Hesperocyparis macrocarpa*) and blue gum eucalyptus trees (*Eucalyptus globulus*). The core overwintering grove is situated 15 meters south of the intersection of Pelton Avenue and Eucalyptus Avenue (Figure 2). Mortality surveys consisted regular searches and collection of dead monarchs encountered in the grove.

On each survey date, we collected all butterfly remnants found within the grove. This included fragments of the monarch's body, wings that had been removed from the body, and bodies of monarch's that were mostly intact. Butterflies collected from each collection date were stored separately. Whole wings/bodies were collected and stored in individual glassine envelopes. Remnants were placed in one of two categories: cache and non-cache. The cache category is for monarchs found in large quantities at a single location. We used this as a proxy for rodent presence, as rodents will cache butterflies, leaving monarch remnants clustered together in one location, usually under organic debris (Figure 3A). The category of non-cache is for butterfly remains found individually within the grove.

We completed mortality surveys at varying intervals multiple times a month during the overwintering period from October 2017 to early April 2018. We targeted some collection times to capture data before and after extreme weather patterns, which can dislodge monarchs from roosts (Brower and Malcom 1991). This resulted in multiple mortality surveys during some weeks. We classified extreme weather events as storms which included strong winds, prolonged periods of cold temperature, and significant precipitation.

In April 2018, we completed necropsies on all monarch remnants collected between November 2017 and early April 2018. For single wings, we tallied the number of left and right forewings and hind wings, as well as the damage and condition. For whole butterflies, we collected data on sex, wing, thorax, abdomen, and head condition to infer the most probable cause of mortality. We only considered whole butterflies in further evaluation of mortality causes. We placed the butterflies into one of four mortality categories: birds, yellowjackets, rodents, or unknown.

Physical damage on whole butterflies was used to infer their cause of mortality. Monarchs missing head and with a hollowed thorax were associated with yellowjacket predation (Figure 3B and 3C). Butterflies that displayed linear wing and abdomen damage resembling handling by bird beak were associated with bird predation (Figure 3D). Monarchs with non-linear wing and body damage were attributed to rodent mortality. Whole monarchs with damage resembling more than one of the possible mortality causes were categorized as unknown. The unknown category also included monarch without signs of physical damage. Some whole monarchs were poorly preserved making it impossible to identify the possible cause of death.

To better understand if rodents were important predators we monitored rodent activity within the grove weekly. We set thirty Sherman traps with a bait of rolled oats and cotton bedding in the evening and recovered on following morning. We set six rows each consisting of five Sherman traps running perpendicular to Pelton Avenue and parallel to Eucalyptus Avenue.

Results

We calculated a minimum estimate of 1,711 dead monarch butterflies between November 2017 and early April 2018. Data compiled by The Xerces Society population count estimates for Lighthouse Field State Beach were 12,000 in November 2017 and 13,533 in January 2018 (Xerces Society). Using the larger January population count yields a mortality estimate of 12.6% for 2017-2018 overwintering season. This number should be treated with caution as it is unknown what percentage of the population turns over due to immigration and emigration during the overwintering period.

Using the entire mortality data set, we found significantly more dead male (mean=50.3, sd=47.6) than female (mean= 39.7, sd=42.0) monarchs on each collection day ($t = 4.8$, p-value < 0.001). Both males and females showed similar temporal trends with the exception of January 26, 2018 when more dead females were encountered (Figure 4). During the month of December where temperature was particularly cold, mortality appeared to be slightly higher in comparison to other months.

Of the 846 whole butterflies we analyzed, we attributed the largest number of the deaths, 49.2% (416) to yellowjackets and 39.2% (332) as unknown (Figure 5). Other sources of mortality included birds (11.0%, 93) and rodents (6%, 5).

Discussion

We estimated a mortality rate of 12.6% for the 2017-2018 overwintering population at Lighthouse Field State Beach with largest sources of predation attributed to yellowjackets 49.2% and unknown causes (39.2%). Birds and rodents also predated on the monarchs, however at a much lower level.

We observed Western yellowjackets (*Vespula pensylvanica*) attacking, killing, and consuming live monarchs in the overwintering grove during the 2017-2018 overwintering period at Lighthouse Field State Beach. We were able to locate two large yellowjackets colonies within 50 meters of the grove in the 2017-2018. One large nest, 50 meters to the east of the grove failed in late December. A second nest, 50 meters to the west of the grove, remained active during the winter and was discovered torn apart on March 29, 2018. Yellowjackets typically hibernate through the winter and become active again once temperatures rise (Wilson et al. 2009). However, yellowjackets remained active in Lighthouse Field throughout the 2017-2018 wintering season. This may be related to relatively warm temperatures and low rainfall in December and January, allowing yellowjackets to persist through the winter and prey upon the monarch overwintering population. The yellowjacket nests were located in open fields with little canopy cover. Here, relatively warm ground temperatures and thermal gain from direct morning sunlight could have enabled yellowjacket to become volant and active earlier in the day. This would allow yellowjackets to prey on monarchs early in the morning when low temperatures within the grove prevented monarchs from being active. Very little literature studies the relationship between yellowjackets wasps and adult monarch butterflies, but Leong et al. (1990) note that wasps are known to attack adult monarchs in California overwintering sites.

Yellowjackets are frequent consumers of human food. A nearby new food service establishment and adjacent school likely provide food subsidizes to yellowjackets in Lighthouse Field. This is an opportunity for outreach and education that will minimize food availability to predatory yellowjackets. Managers could also help protect monarch through a yellowjacket vigilance and control program. This program could be aided by citizen scientists to identify yellowjacket nest locations. There are many simple effective methods for eradicating yellowjacket nests. One low cost non-toxic method is to place dry ice on the nest opening at night then cover with a cloth. As the dry ice becomes gaseous, the CO2 sinks into the yellowjacket nest, killing adults and pupae. We recommend managers employ a citizen science outreach program to educate people about the importance of a tight waste stream and to identify yellow jacket nest locations. This should be followed by removal of yellow jacket colonies in Lighthouse Field. We also recommend placement of traps in the surrounding landscape to assess abundance of yellow jackets.

Peromyscus spp. are widely distributed rodents that are common in monarch overwintering sites and are known to consume the bodies of the monarch but not the wings (Davies et al. 2008). Glenndinning and Brower (1990) studied five species of mice in Mexico finding that between the months of December and late February, monarchs are increasingly susceptible to foraging mice when low temperatures cause monarchs to fall to the ground and render them flightless and unable to return to their clusters. They found *Peromyscus melanotis* largely utilized monarchs as a food source and had high reproductive rates during the monarch overwintering season.

Survey work at Lighthouse Field during 2016-2017 found large numbers of monarch wings in rodent caches (Henry pers comms). While the mortality source could not definitively be attributed to rodents, the caches implicated rodents as potential predators. In 2017-2018 rodents did emerge as an important monarch predator based on necropsies. This may be due to low rodent populations at the site during the 2017-2018 season. We had low capture rates within the grove with the exception of days with significant precipitation. This could be related to rodents seeking extra shelter inside the Sherman traps to protect themselves from precipitation. Another explanation for elevated rodent capture rates during precipitation events may be related to the combined effects of increased surface activity in response to the presence of water and the increased presence of flightless monarchs on the grove floor. Restoration efforts may help suppress rodent populations through active vegetation and woody debris management.

Cold temperatures (<55ºF) and moisture from precipitation or high relative humidity and heavy dew can render monarch nonvolant (Brower and Calvert 1985). Inactivity can increase mortality rates (Alonso-Mejia and Arellano-Guillermo 1992). Cold weather leaves monarchs vulnerable to predators such as yellowjackets and birds. Furthermore, Brower and Calvert (1985) suggest that colder temperatures increase bird activity, as they need to feed more during cold periods.

Recommendations

Future studies on Lighthouse Field State Beach overwintering population should provide comparative datasets for 2017-2018. This should include mortality studies for subsequent years and analyses of archived monarchs collected during the 2016-2017 overwintering season. Ideally, mortality surveys should occur at short regular intervals (1-4 days) to better understand factors (i.e. environmental conditions) that influence mortality during the overwintering season.

Further research on the relationship between monarchs and yellowjackets should be pursued. Our work shows that the monarch overwintering mortality rate can be significantly decreased by immediately eradicating yellowjackets nests. The efficacy of this management action could be reassessed with 2018-2019 mortality surveys and subsequent comparison to our 2017-2018 mortality data.

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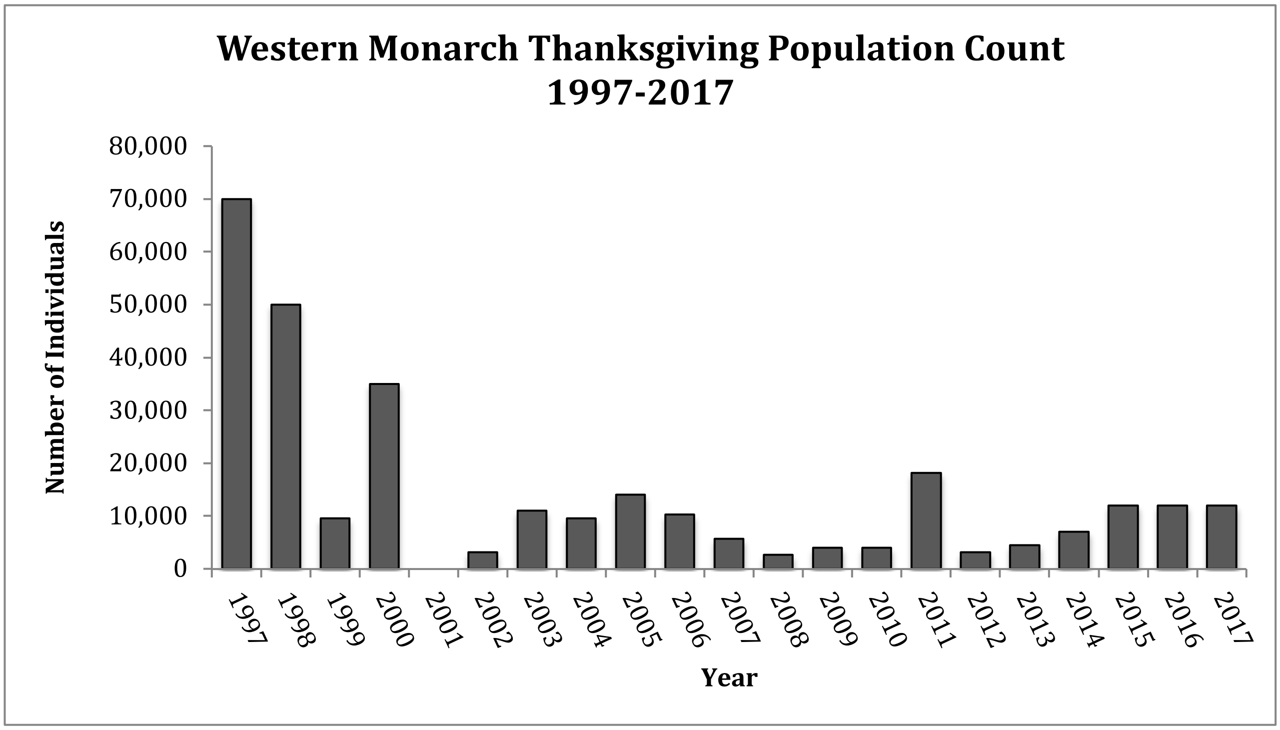

Figure 1: Monarch population size at Lighthouse Field State Beach over the past 20 years. Data were collected from citizen scientists during the month of November in each of the years. Data from 2001 are unavailable. (Western Monarch Count Resource Center, 2018)

Figure 2: Lighthouse Field State Beach overwintering grove. Search area outlined in blue.

Figure 3A: Cache of butterflies indicating possible rodent predation.

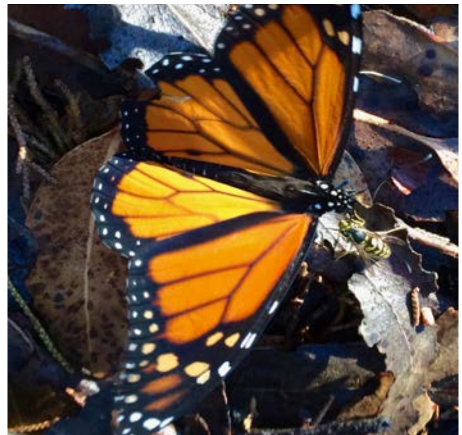

Figure 3B: Yellowjacket wasp attacking monarch on the ground of the grove.

Figure 3C: Dead monarch with a missing head and hollowed thorax, a possible indication of yellow jacket predation.

Figure 3D: Monarch displaying damage to both rear wings and the abdomen indicating possible bird predation.

Figure 3: Photos depicting rodent, yellowjacket, and bird predation. (Source: Groundswell Coastal Ecology)

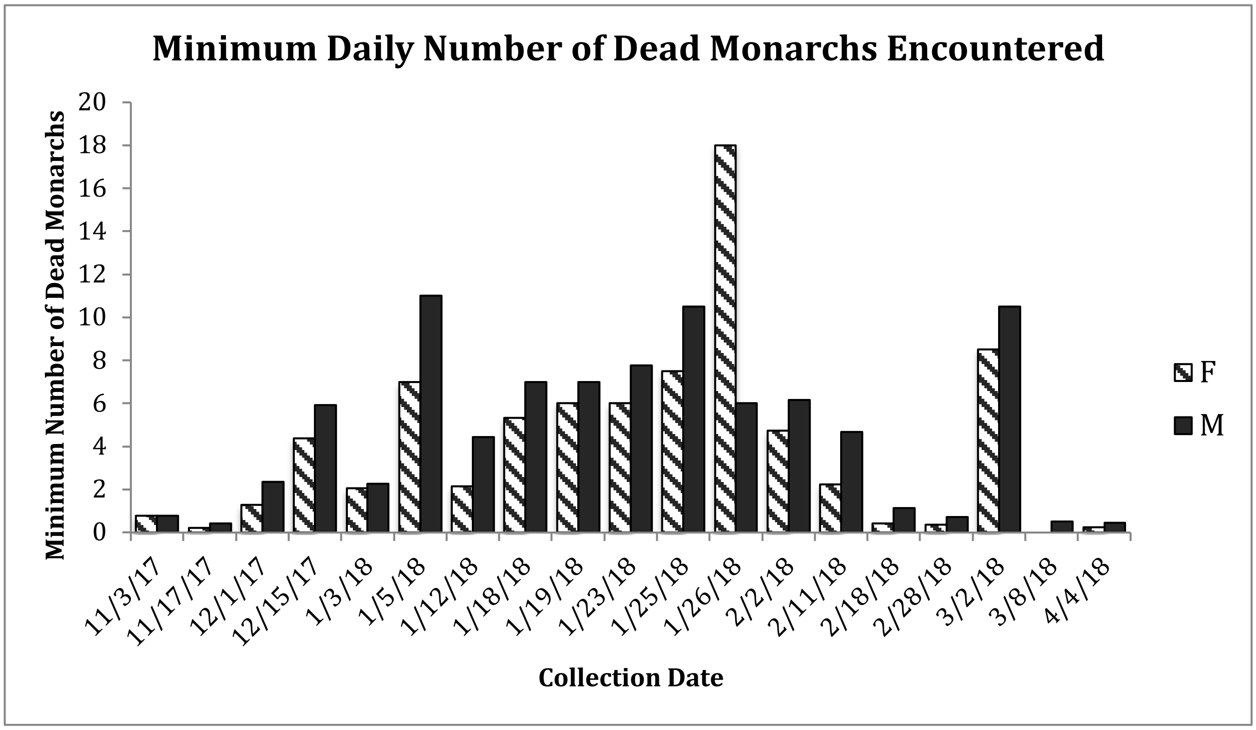

Figure 4: Minimum daily number of dead monarchs by sex between November 2017 and March 2018. Values were converted to daily values to correct for variation in the intervals between each collection date.

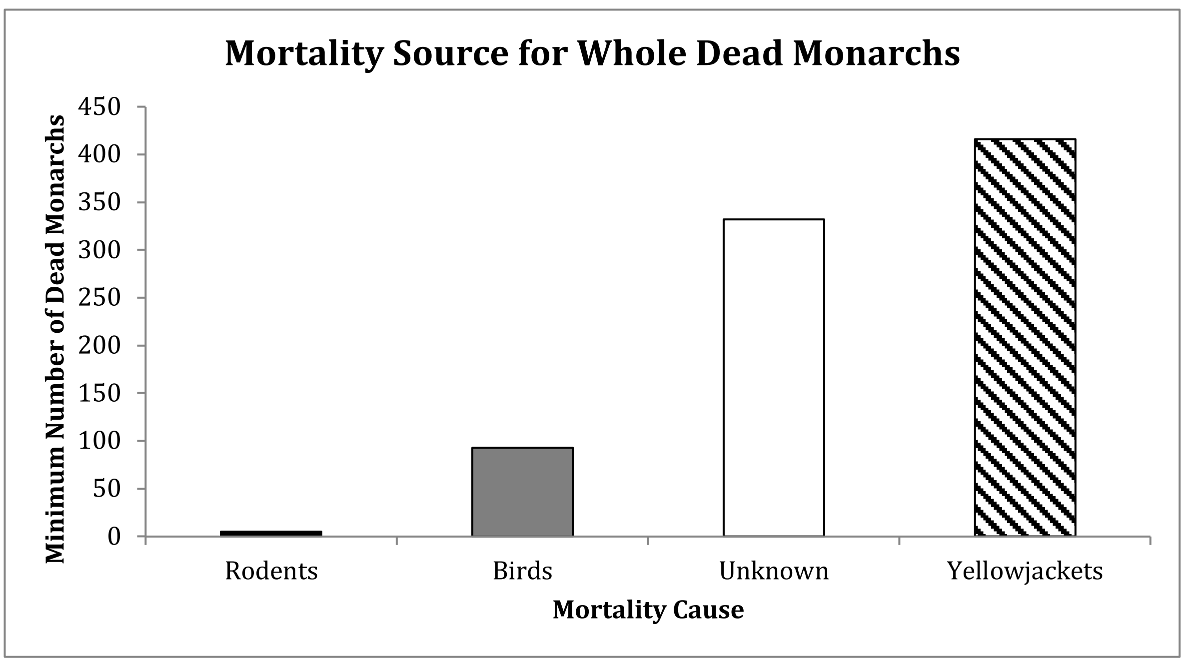

Figure 5: Minimum number of dead whole monarchs encountered for each mortality source.