HOW DOES TEMPERATURE AFFECT THE EMERGENCE AND PEAK ABUNDANCE OF ADULT TAYLOR CHECKERSPOT BUTTERFLIES (*EUPHYDRYAS EDITHA TAYLORI*)?

by

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Abstract

How Does Temperature Affect the Emergence and Peak Abundance of Adult Taylor Checkerspot Butterflies (*Euphydryas Editha taylori*)?

Alicia Netter

Taylor's checkerspot butterfly (TCB) is an endangered species that Washington Department of Fish and Wildlife (WDFW) has been monitoring since 2007. Through their partnership with the Oregon Zoo and the Sustainability in Prisons Project (SPP) at the Evergreen State College, WDFW has helped create new populations with captive rearing and release programs. However, monitoring these new TCB populations is often restricted due to time and labor restraints. Although monitoring has been nearly continuous from 2010 through 2023 in the Joint Base Lewis-McChord Range 50 (JBLM R50) and Scatter Creek Wildlife Area (SCWA) locations, there are years where there are low observation counts and/or the first flight or peak abundance is missed. The purpose of this research was to determine whether or not the Julian day or temperature in the form of Growing Degree Days (GDD) can predict TCB first flight or peak abundance. Additionally, since there was low count data, Generalized Additive Models (GAM) were used to estimate peak abundance. However, this study could not find a direct correlation between Julian day or GDD with first flight or peak abundance. This means temperature does not appear to be the main driving factor determining when adult TCB take flight and more research needs to be done to help monitoring and conservation efforts.

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Acknowledgements

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Introduction

Lepidoptera, which includes butterflies and moths, make up one of the most diverse and identifiable insects in the insect order (Powell, 2009). Many species of butterfly act as indicator species for ecosystem health due to their low tolerance to change in their environment. However, globally many butterfly species are in decline (Forister et al., 2021; Sánchez-Bayo & Wyckhuys, 2019). Butterflies are more vulnerable to habitat loss and fragmentation as most species susceptible to temperature changes and can fly only short distances to find new suitable habitats when theirs is disturbed or destroyed. Thus, the presence/or lack thereof of certain butterfly species in a given location can be a great indicator of whether the habitat is healthy or not.

The Taylor's checkerspot butterfly (*Euphydryas editha taylori*) (TCB) was once widespread and located across the vast prairies on the west side of the Cascade Mountains. However, due to colonization and habitat loss, the species is now limited to fewer than 15 locations in British Columbia, Oregon, and Washington (66 FR 54808, USFWS, 2022, Chappell et al., 2001). TCB were listed as an endangered species by Washington state in 2006 and federally protected in 2013 (USFWS, 2013). Early TCB conservation efforts relied primarily on information on the Bay checkerspot (*E. e. bayensis*), which is a more abundant and studied relative to the TCB (Ehrlich and Hanski, 2004). While captive rearing and release programs have led to more studies on TCB in recent years, there is still a large knowledge gap in their biology and life history.

The TCB has faced many threats over the years. Although a large part of their small population is due to human development, other factors such as the use of pesticides and herbicides, the presence of invasive species, and habitat fragmentation and loss have contributed to their diminishing numbers (USFWS, 2013). TCB are not generalist species and historically have relied on specific host plants such as the harsh paintbrush (*Castilleja hispida*) and golden paintbrush (*C. levisecta*) species to survive (Buckingham et al., 2016; Severns & Stone, 2016). With conifer encroachment and invasive species such as Himalayan blackberry (*Rubus bifrons*) and Scotch broom (*Cytisus scoparius*) taking over prairies, the host plants needed by TCB are choked out (Schultz et al., 2011). As humans combat these invasive species with chemicals, native butterflies and plants are poisoned and killed inadvertently as well (Mallick et al., 2023). While TCB has adapted to use some non-native species as host plants, such as plantains (*Plantago spp.*), these butterflies are not as hardy as the TCB that are able to use native host plants (Dunwiddie et al., 2016). As climate change and invasive species change the landscape, the relationship between TCB and its host plants becomes more important in understanding the survival of the species in the future.

To monitor TCB populations better, the goal of this project is to determine how temperature affects adult butterfly's first flight. This includes the date of first flight and the approximate date of the peak abundance of adult TCB in flight. Flight time is an important monitoring tool as it can indicate changes in population size and response to environmental changes. With climate change making weather and temperatures fluctuate more drastically than historical records, sensitive butterfly species can also act as an indicator species for ecosystems as a whole. Also, as temperatures change, so do insect and plant phenology. Historically, TCB emerge as adults, fly, and lay their eggs between mid-April to mid-June, exactly when their historic host plant, the golden paintbrush, is in bloom (Dunwiddie et al., 2016). These changes in temperature may also change the time at which it takes to accumulate Growing Degree Days (GDD) necessary for the TCB and/or their host plants species to reach certain life cycle milestones (Nufio et al., 2010; Cayton et al., 2015). If changes in temperatures and weather cause their host plant to bloom too late or too early, the TCB might not have an important food source and die or they may not have the visual cues to lay their eggs on these preferred plants (Dunwiddie et al., 2016). Therefore, part of this study is to determine whether or not it is appropriate to use GDD to predict the number of n days it takes for TCB to take their first flight or reach peak abundance.

In addition to using GDD, our goal is to determine if GAM-based models are a good way to predict TCB peak abundance. Monitoring vulnerable species takes a lot of time and resources that biologists do not have, often leading to low count data. GAM modeling can be used to predict non-linear data with low data count and has been demonstrated to be effective in other lepidoptera species monitoring (Dennis et al., 2013; Wepprich et al., 2019). This information will be valuable to the Washington Department of Fish and Wildlife (WDFW) as they can use this information for better monitoring and possible management changes needed to account for climate change temperature shifts.

WDFW has collected data from 2007 through 2023 using parameters based on work by Pollard and Yates done in 1993. Observers went into the field up to three times a week if these conditions were met and continued until the abundance of adult butterfly present reached a near zero number (USFWS, 2022). First flight and peak adult abundance will then be compared to historic weather data for the area(s) and compared to see if there is a correlation.

In summary, the goal of this study is to help WDFW with current monitoring and future management of TCB which is critically endangered. This study will determine whether or not temperatures can accurately determine when butterflies will emerge and if temperatures dictate the duration and peak flight times of TCB. This will better help WDFW in monitoring TCB in the future as they may currently be missing important data with their current monitoring methods. This will also help managers determine the possible effects that climate change might have on the butterflies.

Literature Review

Overview

The Taylor's checkerspot butterfly (*Euphydryas editha taylori*) (TCB) is a victim of habitat loss and degradation. Where once TCB was located across vast oakland prairies on the west side of the Cascade Mountains, the species is now limited to a few isolated populations (66 FR 54808, USFWS, 2022). TCB needs prairies to survive, and yet prairies are amongst one of the rarest habitats in Washington left after colonization (Chappell et al., 2001). Much of what is known about the species has been discovered from the organizations that are responsible for captive rearing and releasing TCB populations (Ehrlich and Hanski, 2004). To monitor TCB populations better, the goal of this project is to determine how temperature affects adult flight. This includes the date of first flight and the approximate date of the peak abundance of adult TCB in flight.

This literature review will go over why the TCB is important and the challenges this endangered species faces. TCB are, in part, endangered because of their specialized needs and their high dependency on a healthy prairie habitat. Prairie habitat and direct threats to TCB are then discussed in detail before diving into their conservation history. Between habitat loss, habitat degradation, pesticides, and herbicides, it is fortunate that the TCB was saved at all when it was finally listed as endangered in Washington state in 2006. Thankfully, the Washington State Department of Fish and Wildlife (WDFW) had the foresight to see TCB decline and started a captive rearing program with the Oregon Zoo in 2004 and partnered with the biologists on JBLM for release sites. Even though their population is now increasing thanks to these programs, TCB still have an uphill battle as the changing weather patterns and temperatures play a key role in their survival. While extreme temperatures can cause a decline in populations, they can also predict what populations can do. Using other species of lepidoptera as examples, this literature review will also cover how temperature, using Growing Degree Days (GDD), can predict different when different life stages will occur within a species. The goal is to use the GDD to see if it can predict first flight and peak flight data on TCB. Knowing this information will help WDFW better monitor their population and it can also forecast how climate change might have an impact on future TCB populations. TCB have limited time as adults and if the plants they need to survive are not in bloom, the population could easily go extinct.

Importance of Butterflies

While there is still much that is unknown about the TCB, butterflies in general play many important roles within their ecosystems. One of those roles is to be a pollinator to a large variety of flowering plants. TCB, along with many of the over 14,500 species of butterflies, are nectivorous (Winfree et al., 2011). While not generally as effective as bees, when butterflies travel from flower to flower searching for nectar they inadvertently spread pollen (Barrios et al., 2016). There are even many mutualistic species of plants and butterfly whose relationships are so co-dependent that they could not exist without one another (Ehrlich and Raven, 1964). While not solely co-dependent, TCB feed on and are hardier when reared with paintbrush species (*Castilleja spp.*)—a plant which until recently was also threatened (Buckingham et al., 2016; USFWS, 2023).

Additionally, TCB and other butterflies are also good indicator species. Indicators are species that, due to their role in a food web, can be the first species to show early signs of changes in the health of an ecosystem (Sampson, 1939; Bakker, 2008). Due to their finicky and fair-weather nature, if TCB are thriving, so is the native habitat. Inversely, if the habitat quality is poor, TCB struggles to adapt to new situations and their population suffers (Linders et al., 2019).

As the climate continues to change, TCB may be indicative of prairie health as temperatures rise in the Pacific Northwest. It can also be an indication that invasive or encroaching species are changing the microclimate and thus negatively impacting the survivability of TCB.

Despite their awful taste, TCB also provide food to many species of animals. TCB absorb iridoid glycosides from paintbrush or plantain species that they feed on during their larval caterpillar life stage (Dunwiddie et al., 2016; Haan et al., 2021). While this deters some predators, birds and non-native European mantids (*Mantis religiosa*) still use the TCB as a food source. Caterpillars that are edible provide a food source that is high in protein and are sought out by many insectivorous bird species (Gauweiler et al., 2022). Generalist and insectivores, such as Steller's jays (*Cyanocitta stelleri*), American crows (*Corvus brachyrhynchos*), American robins (*Turdus migratorius*), and western bluebirds (*Sialia mexicana*), have been seen waiting at TCB sites looking for easy meals after caterpillars are released (Cook, 2023; Linders et al., 2014).

Habitat Needs

TCB need open prairies and grasslands that are surrounded by forested areas in order to survive. Historically, those areas were filled with Oregon white oaks (*Quercus garryana*) as they grow in sub-zones of prairies and forested conifers (Larsen and Morgan, 1998). These areas provide the right soil type needed for the host plants that they are dependent on (USFWS, 2022). However, both Oregon white oaks and prairies are now rare in Washington state and mostly fragmented.

Another reason why TCB are so threatened is because they are not generalists when it comes to food sources and egg laying. They rely on plants in the figwort family (*Scrophulariaceae)* to survive. Eggs laid on harsh paintbrush (*Castilleja hispida*) and golden paintbrush (*Castilleja levisecta*) tend to be hardier than eggs laid on other figworts (Buckingham et al., 2016). However, golden paintbrush was also considered to be endangered until 2023 and paintbrush species are rare or even no longer extant on current TCB population locations (USFWS, 2022; USFWS, 2023). Other native host plants include marsh speedwell (*Veronica scutellata*) and American brooklime (*Veronica beccabunga*). Even in areas where native host plants are still present, isolated sub-populations appear to have developed different preferences for non-native host plants, such as plantain (*Plantago spp.*) and thyme-leaved speedwell (*Veronica serpyllifolia serpyllifolia*), that appear to be related to the quality of the habitat (Severns & Grosboll, 201; Dunwiddie et al., 2016, USFWS, 2022).

Threats

Changes in Habitat

Habitat loss, fragmentation, and degradation are the primary threats to TCB long-term survival. Washington has less than 3% of its historic native prairies (Chappell et al., 2001). This is due in part to urbanization and agricultural growth. Prairies often have fertile soils for crops and provide grass for livestock making them ideal for settlements. These early settlements then lead the way for the construction of railways and, in turn, led to a greater influx of people and more construction on the prairies (Norton, 1979). As prairie habitat began to dwindle and fragment, so too did the TCB populations.

Habitat fragmentation has a greater effect on species that are unable to travel long distances. While species like the monarch butterfly (*Danaus plexippus*) are known to travel thousands of kilometers across generations, the majority of butterfly species only live in their adult stage for a short amount of time and can only travel within their isolated, pocket population range (Chowdhury et al., 2021). In some species of lepidoptera, only the males can fly, as females baring eggs are not capable of liftoff (Berwaerts et al., 2002). Populations of TCB have difficulty dispersing as they are non-migratory and can only fly and estimated 1 to 5 kilometers in their short time as an adult (USFWS, 2022). The majority of TCB fragmented populations are further than 5 kilometers apart, but even in locations that are closer together it is believe that populations rarely mix, making it nearly impossible to maintain healthy genetics without human intervention (Potter, 2016).

As for habitat degradation, this can happen in a few ways. Invasive species, such as Scotch broom (*Cytisus scoparius*) and Himalayan blackberry (*Rubus armeniacus*) can dominate a landscape and change its functionality (Schultz et al., 2011). TCB habitat can also be shrunk by native trees with conifer encroachment. In the past, Native Americans used fires to keep prairies open and free of trees (Peter and Herrington, 2014; Norton, 1979). A large part of their livelihood was dependent on the plants found in prairies and Western Washington would look very different without early human intervention (Norton, 1979). However, in the 1960s there was a big push for fire suppression by the United States Forest Service. This allowed Douglas fir (*Psedotsuga menziesii*) to encroach on already dwindling prairies as their seed bank was no longer being burned off (Norton, 1979). Additionally, after fire suppression practices occurred, fires tended to burn hotter and longer due to years of debris build up. This makes it hard for native seeds to survive and allows for Scotch broom and Himalayan blackberry to disperse to the newly disturbed burn site (Pyne, 2010). These invasives shade out and outcompete native plants and need to be removed to meet TCB living requirements. Often times, official workers and volunteers work together to hand pull weeds to better ensure that the plants are killed back to the root, but this takes a lot of time and manpower (Fyson and Bland, 2016). Another way to mitigate for invasives and stimulate native growth is by using controlled burns. However, if fires are too early in the season or are too frequent, this too can be damaging to the TCB population. Research is still being done to find a happy medium with using controlled burns on TCB habitat in particular (USFWS, 2022).

The composition of prairie plants has changed drastically since urban development and the frequency of controlled burns has changed. In addition to invasive species, non-native/naturalized species have also been introduced thanks to human disturbance. Since TCB was first documented as their own subspecies in 1888, they have been reported to use non-native plant species to survive. It is believed that the primary native plants food sources that TCB historical consumed have either been extradited from their habitats or senesce at different times due to human change of the environment (Buckingham et al., 2016). While this adaptation can be useful when other native species are not available, it can also be problematic.

In particular, certain sub-populations of TCB have come to rely on English plantain (*Plantago lanceolata*) as an early food source because they have leaf foliage available in January when larva exit their winter diapause and start to feed (Severns and Guzman-Martinez, 2021). However, sometime in the early 2000s, a fungal pathogen, *Pyrenopeziza plantaginis*, was discovered in Washington and Oregon that causes the leaves of English plantain to die in early winter, leaving a food shortage for caterpillars that have limited mobility to find other food sources (Potters, 2016; Severns and Guzman-Martinez, 2021). This fungal disease also serves as a biological trap as English plantain are able to persist even with the disease and are asymptomatic during the late spring and early summer in which adults TCB are laying their eggs (Severns and Stone, 2016).

Pesticides

Another threat to TCB is the use of pesticides. In 1869, the invasive European spongy moth (*Lymantria dispar*) was introduced into Massachusetts and has since spread and become a permanent pest across North America (Liebhold et al., 1989). In areas where they establish themselves, spongey moths cause massive tree damage and can cause the destruction of entire forests (Liebhold et al., 1992). In fear of losing the timber and tourist industry, the Washington State Department of Agriculture (WSDA) began to spray for spongy moths after they first started making their appearance in the state in 1974 (WSDA, 2013). In most circumstances, the bacteria *Bacillus thuringiensis* var. *kurstaki* (B.t.k.) is used in the pesticide spray it is relatively inexpensive. However, the problem with B.t.k. is that—besides being an irritant to humans and other wildlife—it is a blanket strategy that attacks the gut of any lepidoptera species. This means that non-target butterflies and moths that consume plants sprayed with B.t.k. are inadvertently killed.

There is a safer pesticide that uses the genetically modified nucleopolyhedrosis virus (LdNPV) that specifically targets Tussox moths (*Lymantriids sp*.) which includes species of spongey moths. This virus causes infected caterpillars to climb to high locations where they die and produce a foul liquid. This liquid, in turn, contains pathogens that allow the virus to spread to other Tussox species (Durkin, 2004). While LdNPV is safe for other lepidoptera species, wildlife, and humans, it is extremely expensive to produce and thus only used in known sensitive locations (D'Amico et al., 1999).

In healthy populations, either pesticide would ideally destroy the budding spongey moth population and any native moth/butterfly species that were destroyed would be replaced by populations outside of the spray area. However, because TCB are so specialized, fragmented, and non-migratory, this spray also has been suspected to have affected their population in the long run (Vaughan & Black, 2002).

Herbicides

As for herbicides, generic, all-purpose glyphosate-based herbicide applications, such as Roundup®, can kill most broadleaf plants (Cox, 1998). Spraying whole fields to save time and cost can feel like the only way to save a prairie filled with invasive species. While intentions of removing invasives are good, the generalized nature of these herbicides also kill the native and naturalized plants necessary for TCB to survive. Even spot spraying, while more time consuming and less damaging to the environment as a whole, can impact caterpillars. Like many lepidoptera species, TCB burrow underground to pupate, so even if visible caterpillars are avoided when spraying, those lying dormant can lay in deadly chemicals for long periods of time. In areas where butterfly pupa are exposed to glyphosate, adult emergence is diminished in numbers and delayed in time (Mallick et al., 2023). This can possibly be detrimental to species that are not generalists and have co-dependency on specific host plants. Additionally, even if host plants survive and insects are not directly exposed to herbicides, glyphosate has the potential to change the nutritional value of plants. Certain species of caterpillars that ingest these plants tend to be smaller in size, have slower growth rates, and overall appear to be less hardy if they survive at all (Bohnenblust et al., 2013; Mallick et al., 2023).

Conservation

The *Euphydryas editha* species was first documented by western science in 1852 and the subspecies, *E. e. taylori*, was first documented in 1888 (Shepard and Guppy, 2001; Pelham, 2008). The TCB was so prevalent in the Puget Sound Basin that it was also dubbed the

"Whulge" checkerspot butterfly from the Salish word for the area (Stinson, 2005). By 2001, this subspecies was first listed as a candidate species for the endangered list when it was documented that, of the 50 locations where TCB had previously been found, populations remained in only 15 (66 FR 54808). Stinson (2005) noted that populations had been experiencing extirpation, particularly within the Puget Lowlands of Washington state, and could only be found on one location on Joint Base Fort Lewis-McChord. Washington has also listed TCB as a Species of Greatest Conservation Need (SGCN) under the State Wildlife Action Plan (SWAP) and Priority Species under Priority Habitat and Species program (PHS) (Larsen et al., 1995; WDFW, 2015). Despite all this, it took until 2006 for TCB to be listed as endangered at the state level and federally in 2013 (USFWS, 2013). Thankfully, WDFW did not want to see this species go extinct so captive rearing and reintroduction programs were started in 2004 (Linders, 2007).

In partnership with WDFW, the Oregon Zoo created a captive-rearing program in 2004 with the ultimate goal to release TCB in protected historic prairies across Washington (Linders, 2007). By 2006, the Oregon Zoo had published their rearing methods and had released their first batch of TCB onto several relocation sites by 2007. Their efforts were later helped with cooperation with the Sustainability in Prisons Project (SPP) in 2011. Two of the earliest reintroduction release locations included JBLM Artillery Range 50 (JBLM R50) (2009-2011) and Scatter Creek Wildlife Area (SCWA) (2007 – 2014; 2016). Releases of captive reared TCB also occurred at other locations, however, due to poor conditions or unknown reasons, some of these reintroduction sites failed or continue to have low population counts (Linders, 2007; Linders et al., 2014; Linders et al., 2019).

Current Monitoring Methods

WDFW has collected data from 2007 through 2023. Locations were visited between April and June when the wind was <10 mph and ambient temperatures were ≥11.7 °C (53.0 °F) on days with shadows present or is >15.5 °C (60.0 °F) when there were no shadows present. This was based on work by Pollard and Yates (1993). Observers went into the field up to three times a week if these conditions were met and continued until the abundance of adult butterfly present reached a near zero number (USFWS, 2022). Counting individuals across entire habitat was not feasible due to the large areas, so locations were divided up into transects. The total number of and the size of the transects varied from location to location, however, they did not vary from year to year so there are no sample size errors on a given site in that regard. Observers would count the number of adult TCB within their given transect(s) from a distance to not disturb them. First flight was considered to be captured in years where a near zero number was recorded at the beginning of observations for that given year or in years were a zero count was recorded a less than a few days prior.

Growing Degree Days

Growing Degree Days (GDD) is a measurement of the cumulative number of days when the temperature is within a certain threshold. This can be used to predict the growth rate of plants or animals. Starting from January 1st, the number of days where temperatures fall within that threshold are counted and once it reaches n number of days, a new life cycle is likely to occur. For example, tropical monarch butterflies have a known temperature threshold when growth can occur with a minimum temperature of 11.5 °C to a maximum temperature of 33 °C (Cayton et al., 2015; Zalucki, 1982). When the threshold of the animal is unknown, the agricultural standard is to use 10 °C for the minimum growing temperature and 30 °C for the maximum (Nufio et al., 2010). GDD has been used to predict life cycle stages of other butterfly species. The research done by Cayton et al. (2015) shows that using the agricultural GDD standard can be used to be accurately predict the emergence of butterflies of a large variety of species.

The ability to predict when insect species reach certain life stages is an invaluable tool for management purposes. The USA National Phenology Network (USA-NPN) provides nationwide maps on emergence events of major invasive pest species (USA-NPN, 2024). These maps are created using GDD that are tailored to the specific species. With this knowledge, agricultural practices can be put into place at the right time and/or life stage to protect crops (Crimmins et al., 2020). For example, the best time to use pesticides to kill invasive spongy moth is during their caterpillar stage. The spongy moth caterpillar life stage emergence occurs at 571 GDD (Russo et al., 1993). Since different states reach this GDD at different calendar dates due to natural temperature differences, each state can determine when it is best to spray based on their own GDD using this predictor.

Likewise, GDD can predict when adult butterflies will emerge. Knowing emergence times is important for managing imperiled species for several reasons. When TCB or other insects exist on locations that require lawn maintenance, it may be important to know when adult butterflies emerge so that mowing schedules can be put on hold until the adults have died off. This has worked well for other species of insects. Mowing can replicate natural grazing, can reduce fire fuel, and it can be beneficial to butterfly survival. However, if done at inopportune times it can be detrimental, either killing necessary plants to sustain life or killing TCB directly (Potter, 2016).

Using Generalized Additive Model (GAM) for Phenotype Predictions

A generalized additive model (GAM) is a model method used to predict non-linear, semi-linear, and nonparametric data by smoothing out scatter plots. It changes a generalized linear model (GLM) to an additive one by changing the predictor formula (Hastie and Tibshirani, 1986). This change is important when working with data with low observation counts and for predicting non-linear trends, particularly in nature when population data can vary annually and within the year itself (Guisan et al., 2002; Dennis et al., 2013). GAM-based approaches have been used to predict butterfly abundance and phenological trends even when the number of observations were small (Dennis et al., 2013; Wepprich et al., 2019). Edwards et al. (2023) also recently created a new technique using GAMs specifically designed for butterflies. When calculating abundance over a season, Edwards et al. (2023) used artificial anchor zeros 20 Julian days before and after the first and last recorded observation dates, respectively, to smooth out the GAM model curve.

Climate Change Temperature Implications

As temperatures change, so do insect and plant phenology. Historically, TCB emerge from their pupa state as adults, fly, and lay their eggs between mid-April to early June, exactly when their historic host plant, the golden paintbrush, is in bloom. If temperatures and weather cause their host plant to bloom late or too early, the TCB might not have an important food source and die or they may not have the visual cues to lay their eggs on these preferred plants (Dunwiddie et al., 2016). This may also lead to TCB shifting their preference of host plant to introduced plants, such as plantains, which makes them less hardy even when golden paintbrush is present on site (Buckingham et al., 2016).

Temperature changes may also lead to a higher rate of mortality based on more extreme temperatures on both ends of the spectrum. If extreme cold snaps or heat waves do not directly kill TCB, they could still affect their host plants. Ehrlich et al. (1980) found that in areas affected by droughts, host plants species did not flower or germinate for two or more years which, in turn, caused local decline and extinction for several *Euphydryas* butterfly populations.

Warmer temperatures can also lead to shorter flight windows for certain species of butterflies. In areas that are more sensitive to drought, such as high elevations, the growing season is cut short leading to the decreased duration of the butterfly flight season (Forister et al., 2018).

However, there is some evidence that warmer temperatures may help expand the small amount of prairie and oak habitat in the Pacific Northwest. It is theorized that, because native prairie plants are suited for dryer conditions, they may finally be able to outcompete generalist invasives which thrive at more moderate temperatures and it may slow the rate of conifer encroachment (Bachelet et al., 2011).

Methods

Study Locations

Washington Department of Fish and Wildlife (WDFW) has collected TCB flight data from 2008 through 2023 at several locations. Locations varied by year based on pre-existing known populations, newly established populations, extradited populations, alternative study designs, and availability of observers. The population from Joint Base Lewis-McChord (JBLM) Range 76 is the original extant population from which captive populations were reared. Of the release locations, the JBLM Range 50 (JBLM R50) and Scatter Creek Wildlife Area (SCWA) maintained their populations long enough to be used in the data analysis. Exact longitude and latitude of the study sites are omitted to protect the remaining TCB populations.

Site Characteristics and History

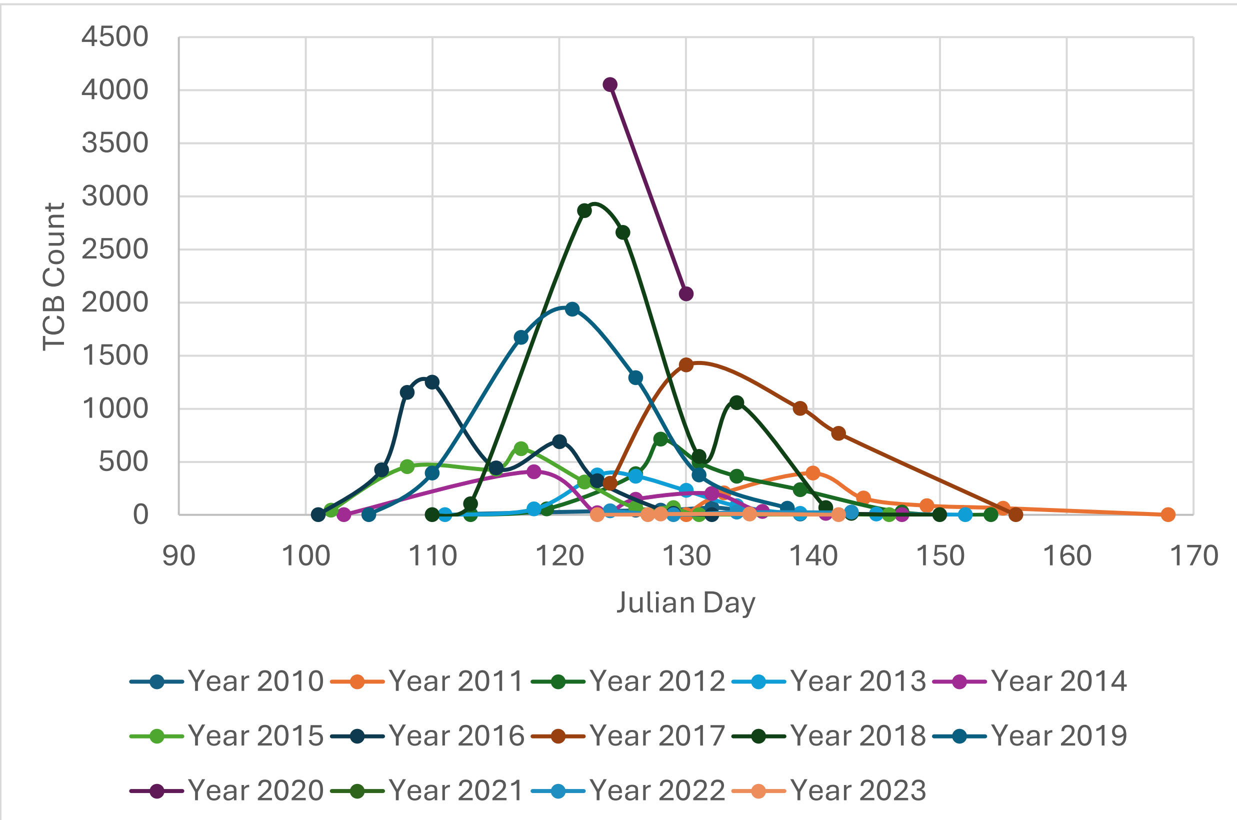
Joint Base Lewis-McChord Range 50 (JBLM R50)

JBLM R50 is a military artillery range is a large prairie that is kept clear and open for practice purposes. This area also has little to no fragmentation and is allowed to experience disturbances (such as fires) that help it maintain a healthy ecosystem. This area was chosen as a reintroduction site because it is similar to the JBLM R76 source population which is also a military artillery range. Captive reared TCB were released at JBLM R50 from 2009 to 2011 and their population has persisted since then. Although the number of days monitored varies from year to year, active monitoring of the location has occurred from 2010 through 2023. Because this location has maintained a healthy population since captive reared TCB were first released, no more re-releases have currently been planned as the population may have reached its carrying capacity (Linders et al., 2015; Linders et al., 2019). Based on the count data provided by

WDFW, there is no apparent pattern in abundance according to year or Julian day, thus the need to examine the effects of temperature (Figure 1).

Figure 1.

JBLM R50 TCB Count Data by Julian Day



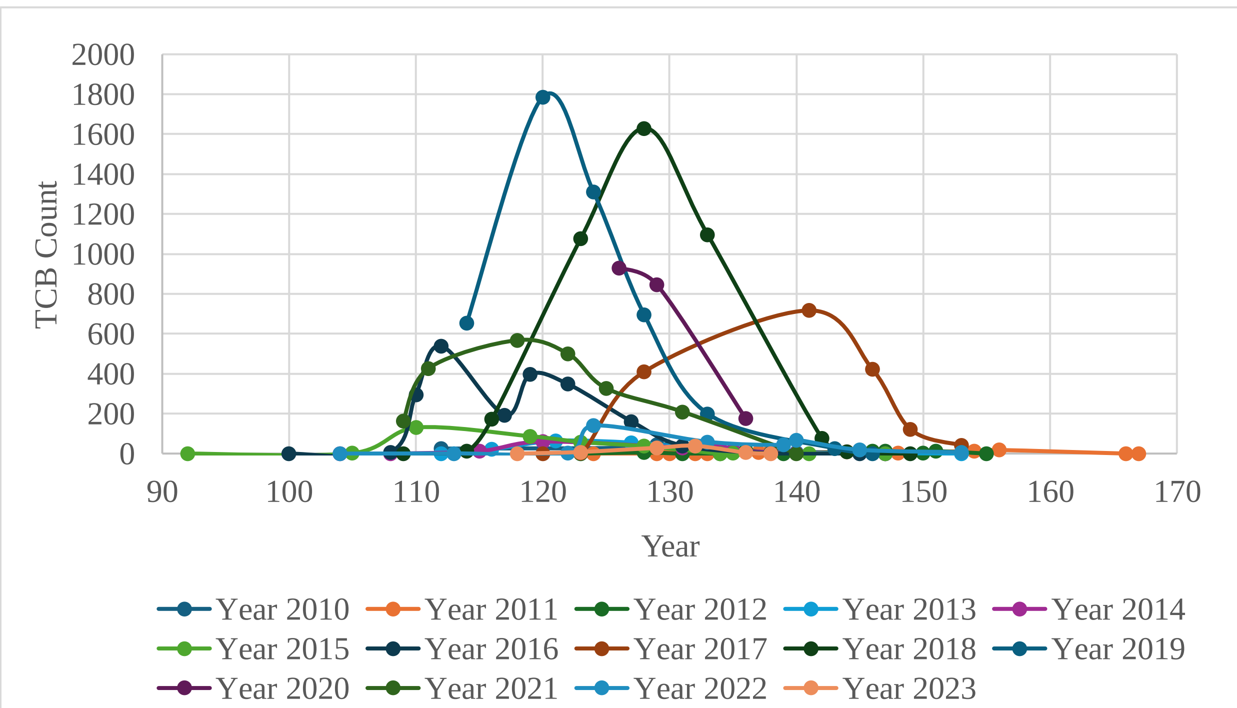
Note. WDFW count data by Julian day of observed adult TCB at the JBLM R50 location from 2010 to 2023. The X-axis starts at Julian Day 90 (March 31st in non-leap years) for ease of display.

Scatter Creek Wildlife Area (SCWA)

SCWA has historically been documented to contain TCB populations and was once considered to be one of the few prime prairie habitats left in western Washington (Dunn & Fleckenstein, 1997). After they went locally extinct, WDFW created a plan to reintroduce captive reared populations in 2007. Although monitoring started in 2008, due to initial poor release conditions, TCB count data from WDFW 2010 through 2023 was used for this analysis (Figure 2). SCWA had TCB captive reared releases continued from 2007 to 2014 and in 2016. Since then, the population has been healthy enough to not require any additional captive releases (Linders et al., 2014; Linders et al., 2019). The population has started to spread and in 2017, SCWA had a spontaneous population in a different field. However peak abundance was low for the majority of its establishment and was practically non-existent by 2023 so it was not included in the analysis (Linders et al., 2019). For the original SCWA location, there is no apparent trend in first flight or peak abundance by Julian day, thus the need for temperature analysis.

Figure 2:

SCWA TCB Count Data by Julian Day



Note. WDFW count data by Julian day of observed adult TCB at SCWA from 2010 to 2023. The X-axis starts at Julian Day 90 (March 31st in non-leap years) for ease of display.

Current Collection Methods

Locations were visited between April and June when the wind was <10 mph and ambient temperatures were ≥11.7 °C (53 °F) on days with shadows present or the temperatures were >15.5 °C (60 °F) when there were no shadows present (USFWS, 2022). This was based on work by Pollard and Yates (1993) on a variety of butterfly species. However, the temperature on location was not recorded and the type of data collected varied slightly by year (i.e. type of shadows present was only recorded from 2019 to 2022 and start and end time for observations was only recorded from 2014 and onward). Observers went into the field up to three times a week if these conditions were met and continued until the abundance of adult butterfly present reached a near zero number (USFWS, 2022). Observers ranged from 2 to 4 people depending on availability and they were trained prior to the beginning of each season (WDFW, 2014). Each habitat was divided into transects and observers counted the number of adult butterflies that were present within their assigned transects for the day/season. While the spectrum of data may have changed slightly depending on project goals for certain years, only the total number of butterflies on a given day at a given location was used in the data analysis. From this data that WDFW provided, estimates for TCB first flight and peak adult abundance for were made.

Temperature Data

Temperature was collected from the historical weather station closest to each site using the dataset available through Visual Crossing Cooperation (VCC), an online database for publicly available historical weather conditions (VCC, 2023). VCC cross references surrounding weather tower data to estimate local historical temperature data. Although the weather stations cannot account for exact microclimate information at each location, it can provide general trends of weather that will be consistent throughout. The weather stations VCC pulled from to create historical weather data for JBLM R50 were KTCM, Tacoma McChord Air Force Base (74206024207), KGRF, Fort Lewis/Gray Army Airfield (KGRF), F6433 Tacoma (F6433), Tacoma Narrows Airport (72793894274), and KTIW. The weather stations VCC used to generate historical weather data for SCWA were Washington stations KE7PBG Rochester (F3822), DW0126 Tenino (D0126), FW8782 Olympia (F8782), Chehalis Centralia Airport (KCLS), Olympia Regional Airport (KOLM), Chehalis (CLSW1), Fort Lewis/Gray Army Airfield (KGRF), and Shelton Sanderson Field (KSHN).

Growing Degree Days

Growing Degree Days (GDD) is a measurement of the accumulated heat (or energy) over a given time period, calculated as the amount of time the temperature in a particular location is above a specified minimum temperature (or sometimes, within a certain range of temperatures). GDD is commonly used to predict the growth rate of crops (Miller et al., 2001) but has also been used to predict life cycle stages of other butterfly species (Cayton et al., 2015). When a specific temperature range or threshold of a species is unknown, the agricultural standard is to use 10 °C for the minimum growing temperature and 30 °C for the maximum (Nufio et al., 2010). Based on this information, GDD was calculated starting from January 1st in a given year using the formula $GDD = ((Temp_{Max}\ °C + Temp_{Min}\ °C)/2) – 10\ °C$. Maximum and minimum temperatures for a given study location were based on VCC weather data and used in the above equation to calculate GDD on any given day, and the year's cumulative total was calculated for each day. When temperatures exceeded 30°C maximum or the 10°C minimum, they were rounded up or down respectfully.

The accumulation of GDD was compared to first observed flight across years to see if GDD can predict when TCB first emerge as adults. This only works for years when the first observation of butterflies is near zero and first flight is assumed to be observed. For certain years, when the first observation was high, it is assumed that the observers missed first flight data. GDD was also be compared to observed peak abundance and a GAM was used (see below) to ask whether GDD is associated with predicted peak abundance.

Generalized Additive Model

A generalized additive model (GAM) can be used to make predictions on phenological outcomes when survey data is sparse and where potentially unknown and potentially non-linear factors may play a role (Wepprich et al., 2019). To see if GAMs were useful to predict peak abundance or smoothing out low count data, GAMs were applied to the JBLM R50 and SCWA locations. GAMs can be used with variations in the gamma number. The standard is run with gamma equal to 1. However, according to Wood (2017), using a gamma equal to 1.4 can smooth out model curves and can reduce the chance of overfitting models. Additionally, anchor zeros can be placed to smooth out scatter plots and models with low data counts (Edwards et al., 2023). Anchor zeros are when artificial zero observation is placed both 20 days before and after the first and last observations, respectively. This not only captures true zero observations, but it also increases the k value. K values need to be greater than or equal to 3 to operate, so this allowed data analysis on years where only 4 or 5 days (n) were monitored (Edwards et al., 2023).

GAM models were created separately using 1) Julian day or 2) accumulated GDD to see if either could predict the peak abundance of adult TCB in flight. Julian day was used to see if there were any unforeseen factors driving the date of peak abundance. The next GAM-based models were run using GDD so that each location could account for their unique variations in temperature by year rather than just the actual date.

To see which version of GAM would be most appropriate for the data collected, seven versions of the model were done for run on a single location (JBLM R50). The seven variations on the models run were GDD with gamma = 1, GDD with gamma =1.4, GDD with anchor zeros and a gamma = 1, GDD with anchor zeros and a gamma = 1.4, Julian day with gamma =1, Julian day with gamma = 1.4, and Julian day with anchor zeros and gamma = 1. Based on the GDD model with anchor zeros and gamma = 1.4, there was no need to run a Julian day with anchor zeros and a gamma =1.4.

The differences in the results are compiled into Appendix I and Appendix II. In addition to the Jullian day or GDD associated with the maximum predicted value from the GAM (i.e. the predicted peak abundance), a measure of uncertainty around the estimate calculated using the range of the predictor of butterfly counts (either Julian day or GDD, depending on the GAM) associated with 95% of the predicted peak abundance. Adding anchor zeros had a tendency to skew estimated peaks, meaning that peak abundance was predicted to occur at either an earlier or later GDD date than to be expected based on observed dates. This is especially true in years where the first and/or observation was already documented to be 0. Based on this, anchor zeros were not used in the SCWA location models. Based on the r-squared and p=values, the unaltered data using gamma = 1 created GAM models that more accurately fit the observed estimated peaks, thus only the gamma = 1 was used for the SCWA location. Both GDD and Julian day were still used separately to account for local differences in temperature and overall temperature trends in a given year.

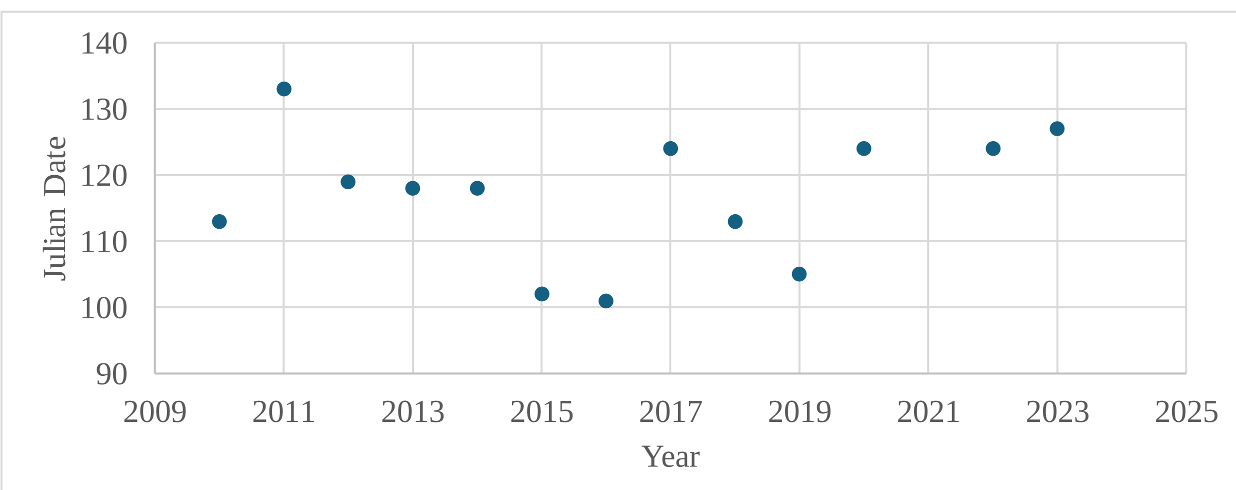
Results

JBLM R50

For the JBLM R50 location, observations occurred from 2010 through 2023. There were minimal observations in 2020 and no observations in 2021 due to COVID-19 restrictions. The first Julian day with a non-zero observation are considered to be the observed first flight of adult TCB (Figure 3). There is no obvious trend over time. The earliest Julian day that a TCB was observed was 105 in the year 2016 with a near zero count of 1 TCB. The latest Julian day TCB were observed taking first flight was 133 in the year 2011. Although 2011 did not a near zero first detection (207 count), a zero count was found on Julian day 130, so first flight is believed to have occurred within the three-day window. This means that first flight was detected between April 14th (Julian day 105) and May 13th (Julian day 133) across all the years sampled.

Figure 3:

JBLM R50 Observed First Flight Date of TCB by Julian Day

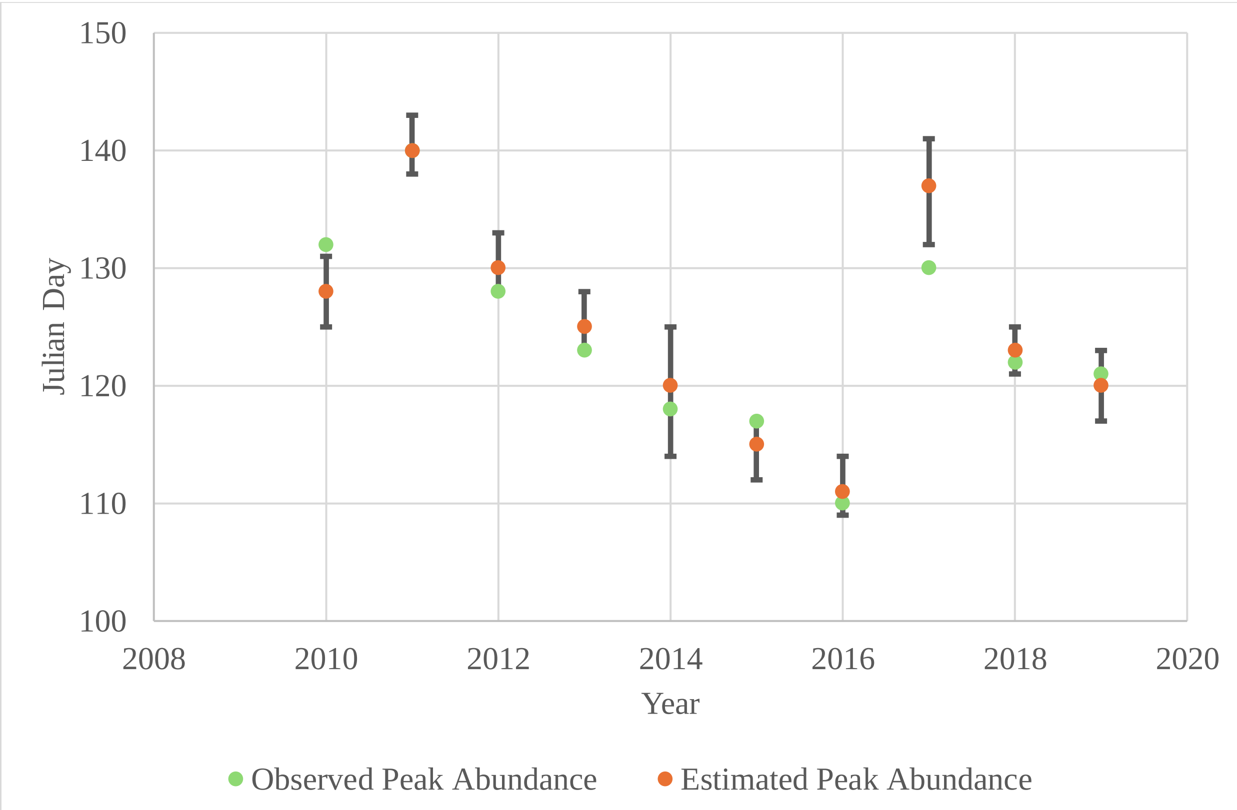


Note. Julian day of the first flight—the first near-zero TCB count—at JBLM R50 from 2010 through 2023 as observed by WDFW. No data was collected in 2021 due to COVID-19 restrictions. The Y-axis starts at Julian day 90 (March 31st in non-leap years) for ease of display.

Julian day was used in GAM models to generate an estimated day of TCB peak abundance which was then compared to observed peak abundance data. Based on modeling trials (Appendix I), gamma =1 was used for the GAM models. Observed peak flight was the Julian day with the highest abundance observed. These observed peaks were also only considered to be true estimated peaks if there was a prior observation with a lower count, thus in 2020 it is unknown if peak abundance was captured. While there was data in 2022 and 2023, their low k values made it impossible to run the GAM model so they were not included in Figure 4. There does not appear to be a consistent predictive relationship between Julian day and either observed or GAM-predicted peak abundance in TCB (Figure 4). The latest peak day is in 2011 on Julian day 140 (May 20th) and the earliest peak day was Julian day 110 (April 21st) in 2016. GAM models fit years where their predicted range overlapped observed peak abundance which were every year except 2010 and 2017. GAM models were more or less aligned with the observed data and did not add any predictive ability for Julian day: Notably, using the GAM predicted peak based on Julian day did not shrink the large month-long window of potential peak abundance.

Figure 4:

JBLM R50 Observed Peak Flight vs Estimated GAM Peak Flight of TCB by Julian Day

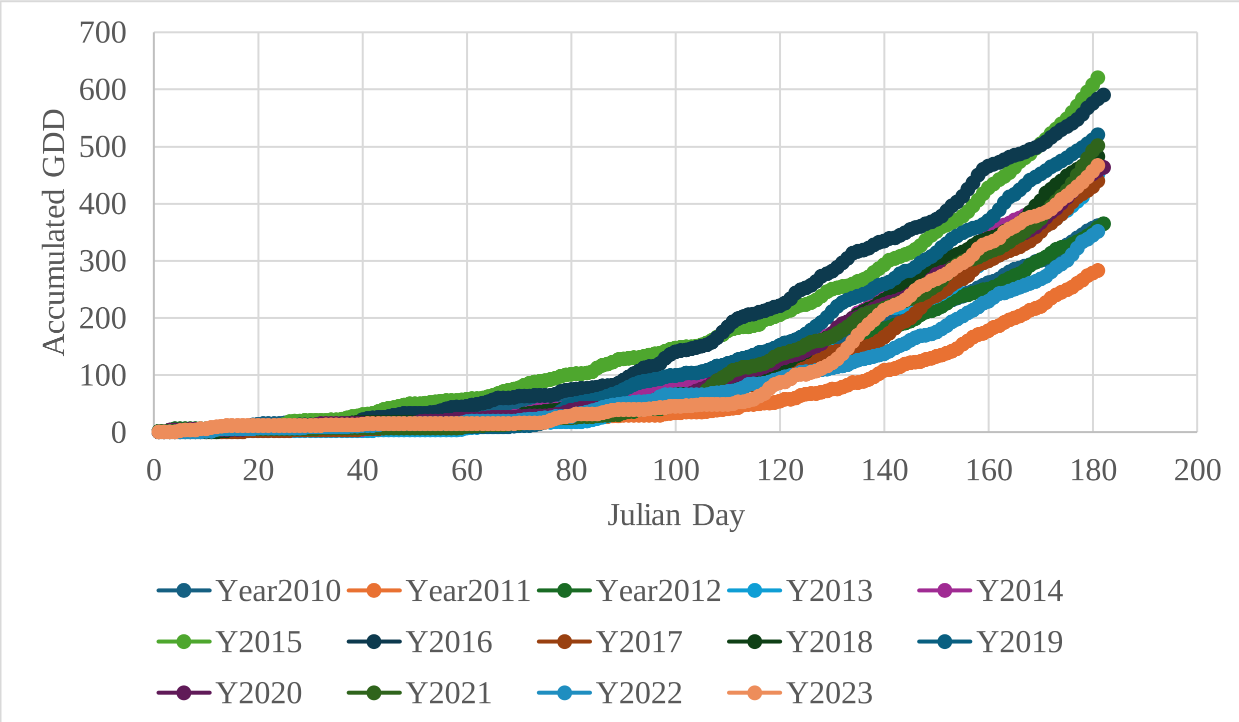


Note. Julian day of peak TCB abundance as observed by WDFW (green) compared to GAM estimated Julian day of peak TCB abundance (orange) at JBLM R50 from 2010 through 2019. The bars on the estimated peak represent the uncertainty calculated for GAM models with gamma = 1 (Appendix I). The Y-axis starts at Julian day 100 (April 10th in non-leap years) for ease of display.

Next, GDD was calculated for the for each year from January 1st through June 30th (Julian day 1 through 180/181 (depending on leap years)) using the temperature data provided by VCC (Figure 4). As expected, accumulated GDD growth rate started to grow exponentially with the warming of the seasons from winter to spring to summer. The year 2011 had the slowest rate of GDD and 2015 and 2016 had the fastest accumulation of GDD (Figure 5).

Figure 5:

JBLM R50 Accumulated GDD by Year

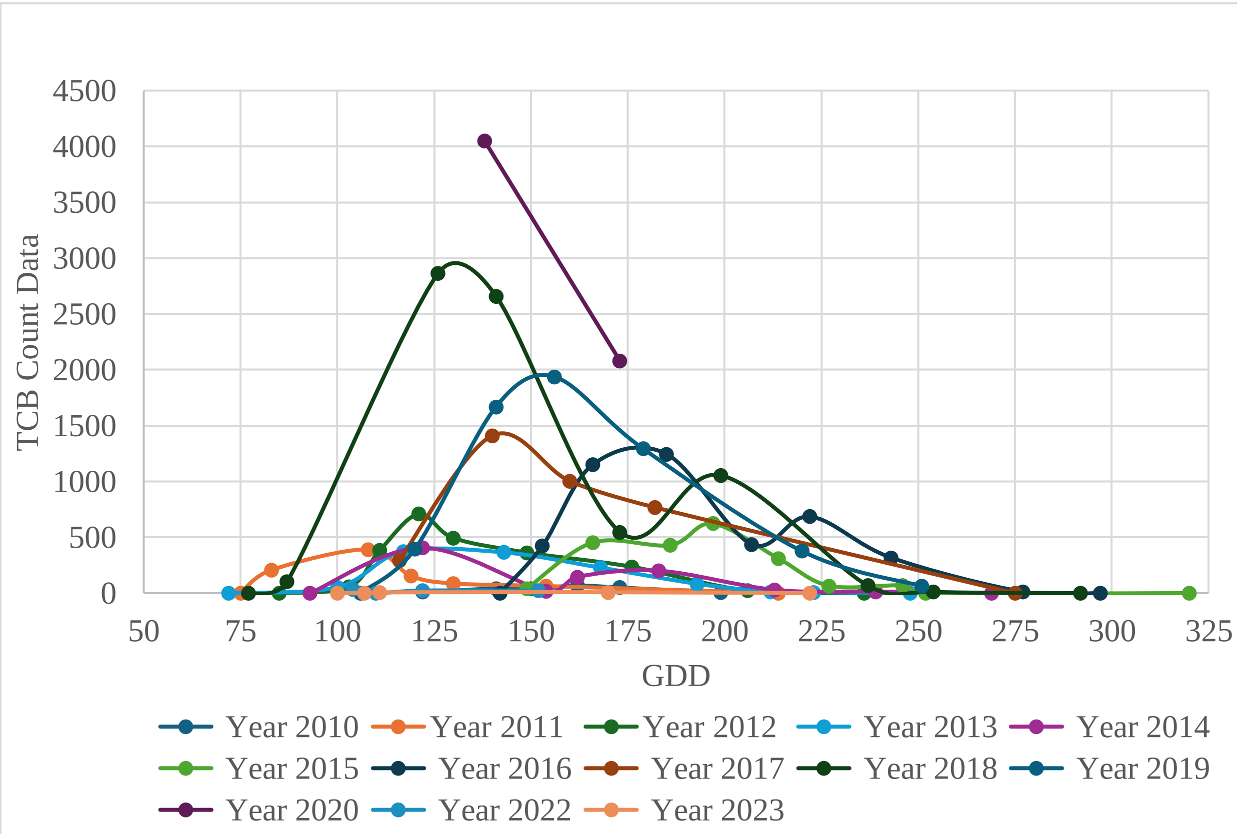


Note. Accumulated GDD by year for JBLM R50 from January 1st through June 31st (1-180/181 Julian days depending on leap years) from 2010 through 2023. Temperature data was provided by VCC.

After GDD was calculated each year, the corresponding count data from WDFW by Julian day was converted and graphed in Figure 6. This data was then used to see if there was a correlation between temperature in both first flight and peak abundance.

Figure 6:

JBLM R50 TCB Count Data by GDD

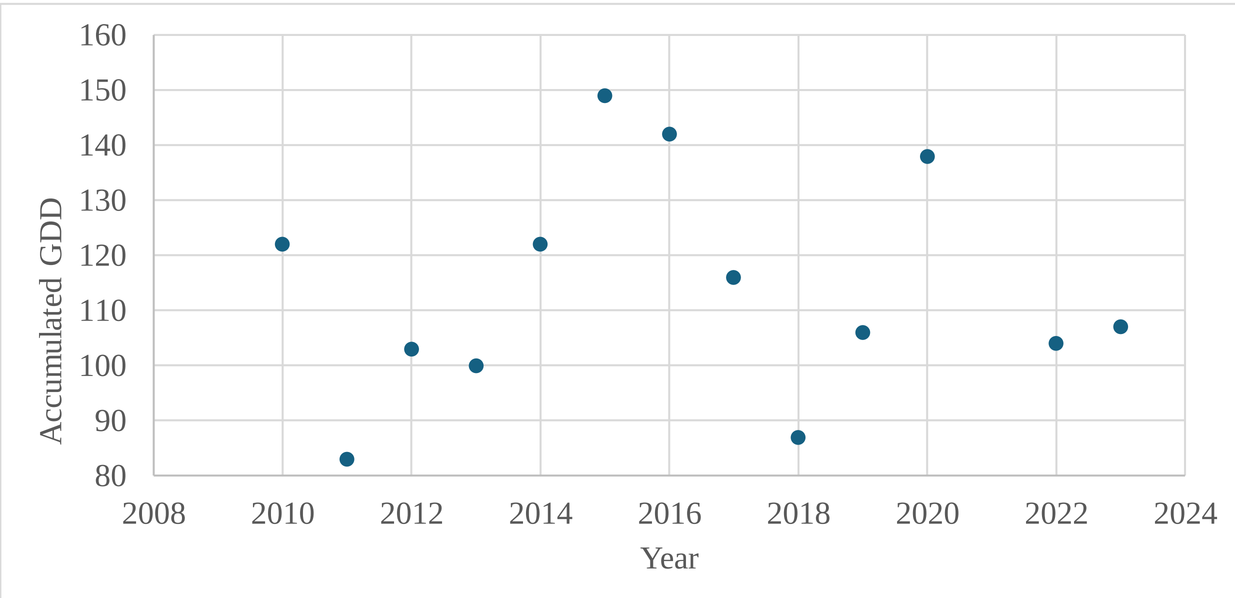


Note. JBLM R50 TCB count data from WDFW by GDD (accumulated GDD on the Julian day of the observation) from 2010 to 2023. There were no observations in 2021 due to COVID-19 restrictions. The X-axis starts at 50 GDD for ease of display.

When first flight by GDD is graphed in isolation, there appears to be no direct correlation between adult TCB emergence and temperature (Figure 7). The earliest observed GDD for first flight was found at 83 GDD in 2011 and the latest was found at 149 GDD in 2015. The first observation date in 2015 had no near zero observations, it is uncertain whether or not first flight was captured this year, however, in 2016 the first flight was recorded at 142 GDD with a near zero observation so it is possible that 2015 still is within the phenotypic range of first flight. This means TCB first flight ranged from 83 GDD to 149 GDD. This large range of GDD makes it difficult to pinpoint a specific date for future observers to start checking TCB.

Figure 7:

JBLM R50 TCB Observed First Flight Date by GDD



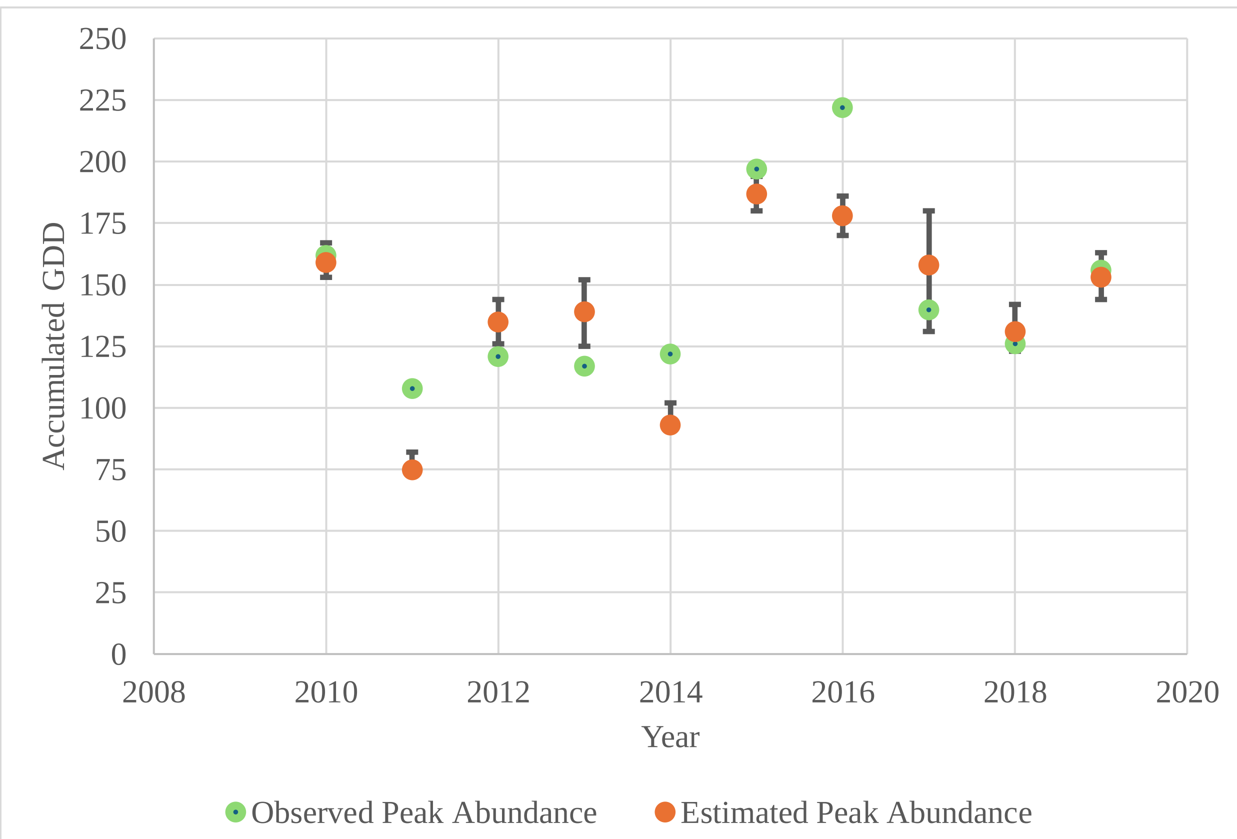
Note. The accumulated GDD for the first observation with a non-zero TCB count by year for the JBLM R50 location. No data was collected in 2021 due to COVID-19 restrictions. The Y-axis starts at 80 GDD for ease of display.

Just like with Julian day peak estimates, peaks for a given year were based on the GDD date with the highest abundance observed with prior lower count data. Thus, in 2020 it is unknown if peak abundance was captured. To predict peak TCB abundance using temperature, GAM models were created each year using GDD. Based on the results in Appendix II, GAM models with gamma = 1 was used. Predicted peaks were compared to the estimated peaks to determine whether or not the GAM model could accurately predict peak abundance in TCB (Figure 8). Like Julian day, GAM models could not be made for 2020, 2022, and 2023 as their k values were too small. It is unclear why 2011 had lower than expected predictions for GAM

GDD models, as the 2011 data did work for Julian day GAM models. In 2014, all GDD GAM models are most likely not accurate as a near zero observation occurred in the middle of the season and returned to a higher count, causing a false second peak; however this did not affect the Julian day predictions. Overall, GAM models are usable for 2010, 2017, 2018, and 2019 as they fit within the model's predicted GDD range for peak abundance.

Figure 8:

JBLM R50 Observed TCB Peak Abundance vs GAM Estimated Peak Abundance



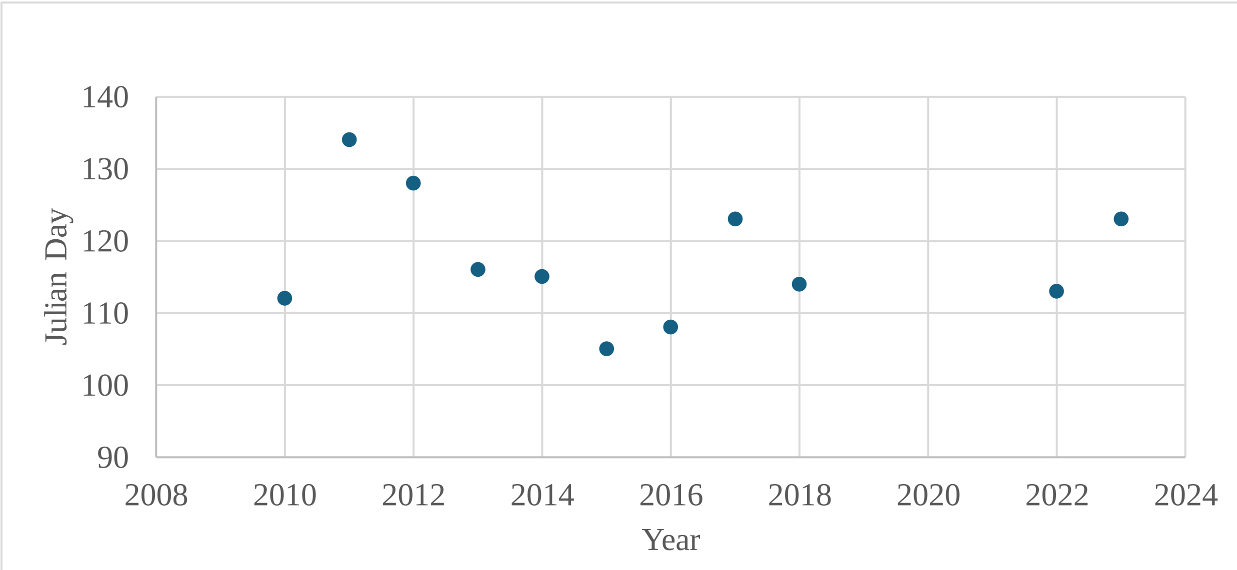
Note. JBLM R50 accumulated GDD at observed peak abundance (green) and at GAM estimated peak abundance (orange) of TCB from 2010 through 2019. The error bars on the estimated peak values represent the uncertainty calculated for the GAM models with gamma = 1 (Appendix II). Peak abundance was not captured in 2020 through 2023 due to low data counts.

SCWA

For the SCWA location, observations also occurred from 2010 through 2023. There were minimal observations in 2020 and no observations in 2021 due to COVID-19 restrictions. The first Julian day with a non-zero observations are considered the to be the observed first flight of adult TCB (Figure 9). Although data was collected in 2019 through 2021, there was no zero count prior to the large count first observations (654 in 2019, 930 in 2020, and 163 in 2021) so there is no way to accurately predict when first flight would have been. Similar to JBLM R50 there is no obvious trend over time. The earliest Julian day that a TCB was observed was 105 in the year 2015 with a near zero count of 2 adult TCB. The latest TCB were observed taking first flight was in 2011 on 134 Julian day with a non-zero count of 6 adult TCB and a zero count the day prior. in the year 2011. This means that first flight was detected between April 14th (105 Julian day) and May 15th (133 Julian day) across all the years sampled. This is only a two day difference from the JBLM R50 location.

Figure 9:

SCWA Julian Day of Observed TCB First Flight

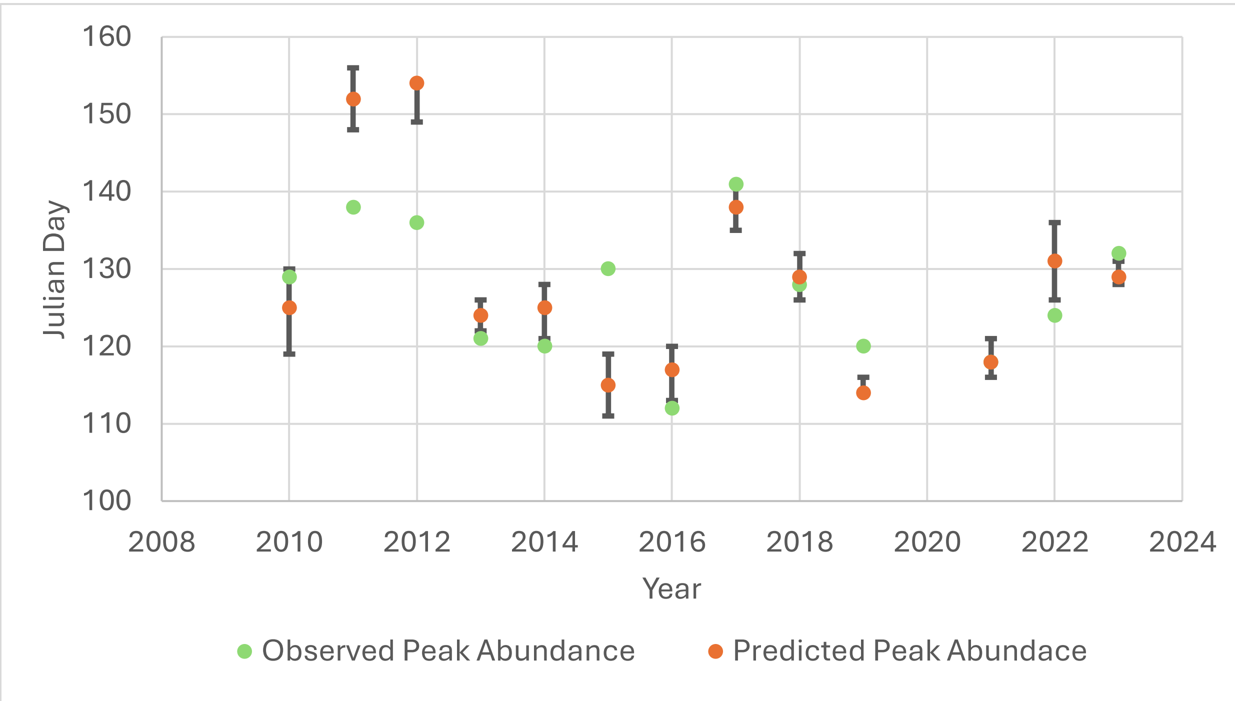


Note. Julian day of TCB first flight at SCWA from 2010 through 2023 as observed by WDFW. Although data was collected from 2019 through 2021, there was no near zero first observation so it is uncertain if first flight was captured. The Y-axis starts at Julian day 90 (March 31st in non-leap years) for ease of display.

Observed peak abundance for SCWA was determined by the highest count number of adult TCB observed by WDFW researchers (Figure 10). Although first flight was not observed in 2019 through 2021, peak abundance is believed to have been captured in 2019 and 2021 because there were observations with lower counts prior to the highest count. Therefore, the only year without an observed year was in 2020 and that was due in part to Covid-19 restrictions and a low data count. 2015 had the earliest peak abundance at Julian day 110 and the latest peak abundance observed on Julian day 156 in 2011 which aligns with what was found in the JBLM R50 location. This window ranged from April 20th (110 Julian day) to June 5th (156 Julian day), which is too large to give recommendations for WDFW on serving peak abundance at the SCWA location. Since there was no apparent pattern of GDD and there was low count data, GAM models were used to see if they could provide a more accurate and smaller window of peak flight times (using gamma = 1).

Figure 10:

SCWA Observed TCB Peak Abundance vs GAM Estimated Peak Abundance by Julian Day

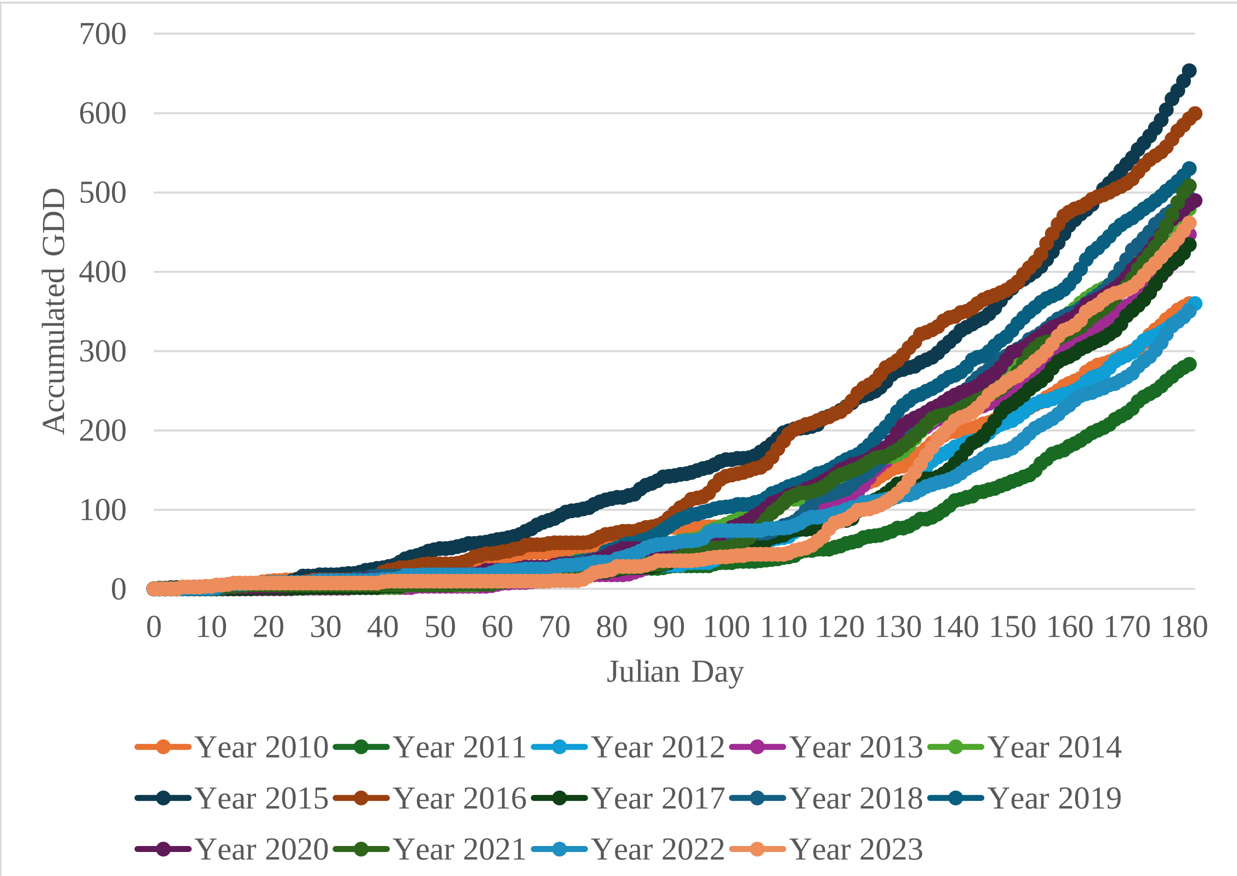


Note. SCWA observed TCB peak abundance by Julian day compared to GAM estimated peak abundance from 2010 through 2023. The error bars of the estimated peak values represent the uncertainty calculated for the GAM models with gamma = 1 (Appendix III). There was not enough data for 2020 due to COVID-19 restrictions.

SCWA had similar temperature trends to that of the JBLM R50 location. The accumulated GDD rates by year were calculated using the VCC weather data (Figure 11). This figure shows that GDD rates in 2011 were the slowest and where the greatest in 2015 and 2016 from January 1st to June 30th. Like JBLM R50, SCWA also saw its slowest rate of GDD 2011 and its fastest in 2015 and 2016.

Figure 11:

SCWA Accumulated GDD by Julian Day

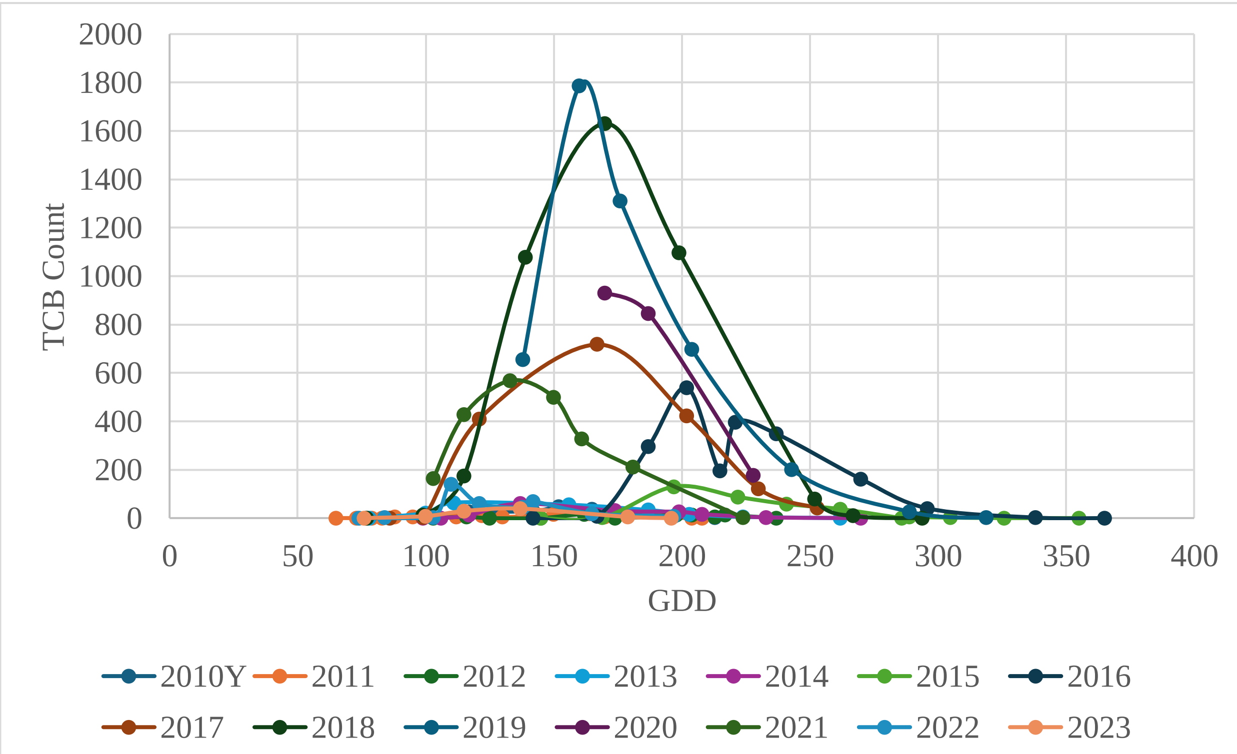


Note. Accumulated GDD by year for SCWA from January 1st through June 31st (1-180/181 Julian days depending on leap years) from 2010 through 2023. Temperature data was provided by VCC.

To determine if temperature affected TCB emergence, count data was converted from Julian days to GDD and graphed (Figure 12). To further break down the analysis, first flight was singled out every year from 2011 through 2023 (Figure 13). Although data was recorded in 2010 and 2019-2021, the first observation for those years was not a near zero number, therefore we cannot say for certain first flight was captured those years. The earliest observed GDD for first flight was found at 84 GDD in 2022 and the latest was found at 169 GDD in 2015. This large range of GDD makes it difficult to pinpoint a specific date for future observers to start checking for first flight at the SCWA location.

Figure 12:

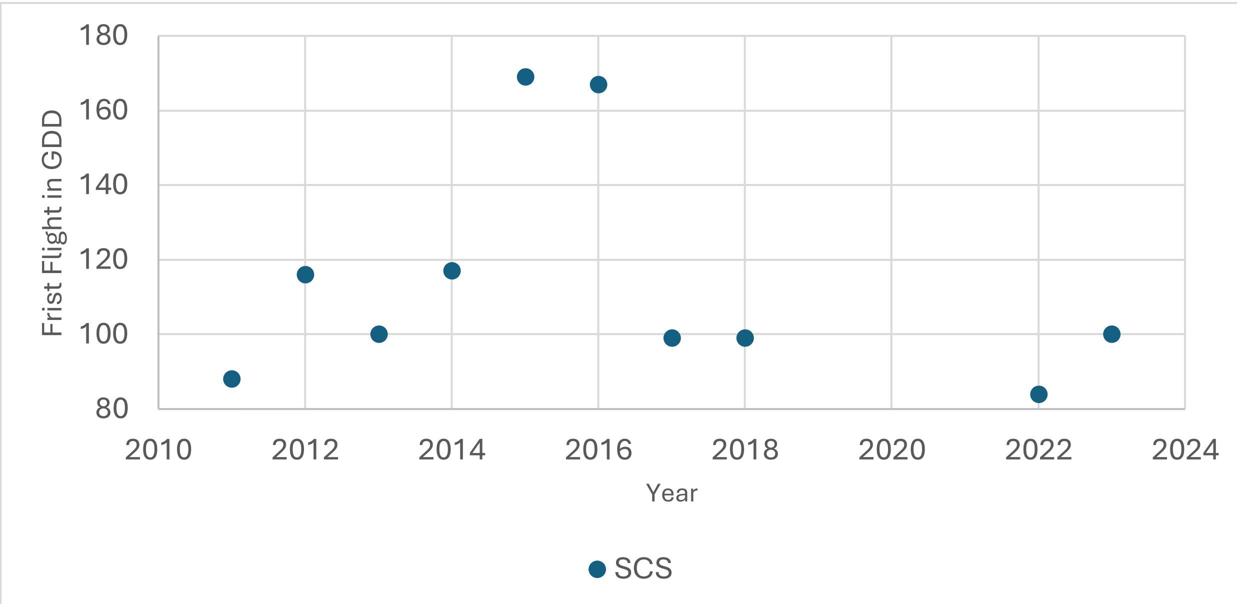
SCWA TCB Count by GDD



Note. SCWA TCB Count Data by GDD (accumulated GDD on the Julian day of the observation) from 2010 through 2023.

Figure 13:

SCWA TCB Observed First Flight

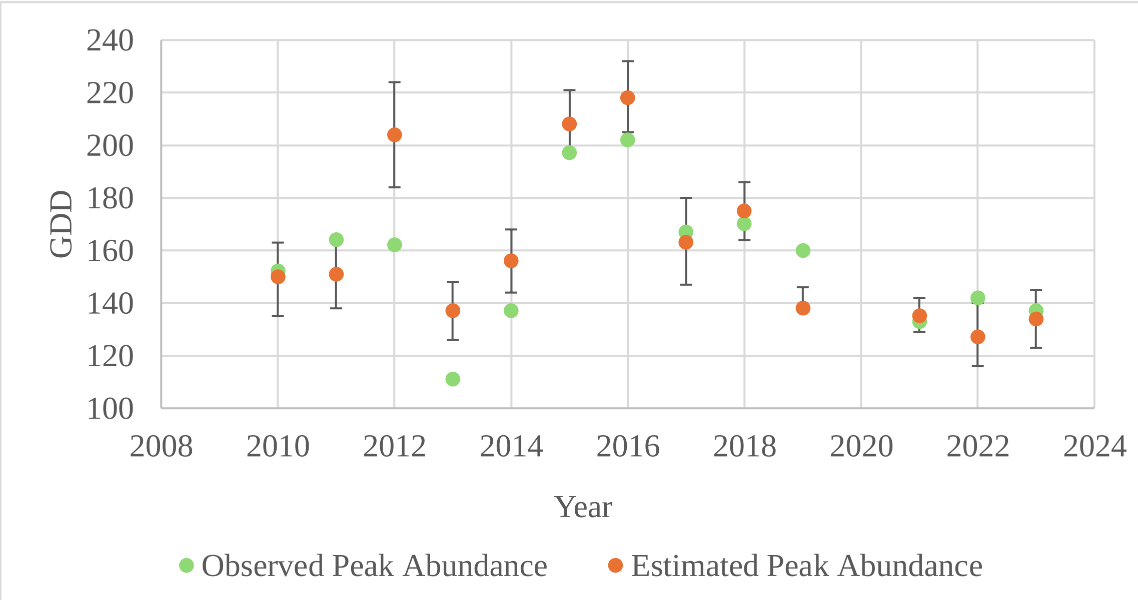


Note. SCWA TCB first flight by GDD from 2010 through 2023. Although data was recorded in 2010 and 2019 through 2021, there were no near zero observations at the beginning of each year, therefore first flight cannot be confirmed. The Y-axis starts at 80 GDD for ease of display.

The observed peak TCB abundance overlaps with the GAM models ranges in years 2010, 2012, 2017, 2018, 2021, and 2023. Regardless of overlap, GAM or observed estimated peaks did not provide a small enough window to make recommendations on when researchers should go out to capture the true peak abundance at the SCWA location. The earliest observed peak abundance was found in 2013 at 111 GDD and the latest was 202 GDD in 2016.

Figure 14:

SCWA TCB Observed Peak Abundance vs GAM Estimated Peak Abundance by GDD



Note. SCWA observed TCB peak abundance (green) compared to GAM estimated peak abundance (orange) from 2010 through 2023. The error bars on the estimated peak values represent the uncertainty calculated for the GAM models with gamma = 1 (Appendix III). Due to COVID-19 restrictions, not enough data was collected in 2020 to definitively claim that peak abundance was captured during observations and there was not a high enough k value to run a GAM model. The Y-axis starts at 100 GDD for ease of display.

Discussion

The purpose of this study was to help WDFW better monitor TCB. With limited time and resources, there were many years and locations that did not receive enough attention to capture first flight or peak abundance. One difficulty that arose with this study was the limited amount of data at given locations. Although WDFW collected data from a total of 10 distinct populations within Washington, many of these died out before trends could be established. Also, some of these locations had fewer days of observations due to either their remote locations or observer availability. For example, TCB data was collected from Peak Maker (PMC) 2012 to 2015, however the first year had its highest count of 21 individual adult TCB, by the following year they had gone locally extinct. COVID-19 restrictions also limited the data WDFW was able to collect in 2020 and 2021. The hope of this study was that, even with low data counts, this study could be used to help time the use of resources more efficiently.

This study could not determine a consistent pattern using Julian day or GDD to make predictions of when TCB would take their first flight. Julian day was used to see if outside factors may be influencing the time of first flight, but there was a large range of dates for both locations, making it difficult to use it as a predictor. Dates for TCB first flight ranged from mid-April to late-May making the difference in years to be over a month at times. It is hard to justify that Julian day is a better predicter than Pollard and Yates (1993) at capturing first flight.

If temperature as measured by GDD was a more consistent driving factor for day of first flight, the expectation would be tighter range of variation in GDD values at first flight (compared to Julian day). For JBLM R50, the range of first flight occurred was 83 GDD to 149 GDD, making a large window of 66 GDD. For SCWA, the range of first flight occurred was 84 GGD to 169 GDD, making it a window of 85 GDD. While this large range of dates cannot inform

WDFW when to start collecting data each year, there were still some interesting findings that should be looked at for future research.

If accumulated temperature was a factor in determining TCB first flight, it would be expected that in colder years, first flight would occur later in the season and in warmer years, first flight would occur earlier in the season. This was found to be true as in 2011 and 2022, the years which had the lower accumulated GDD, TCB had some of the latest emergences in mid to late May. Conversely, in 2015 and 2016, the years which had the highest accumulated GDD for both locations, TCB emerged the earliest in mid-April. While temperature certainly plays a role in emergence, if it were the sole factor, the phenotypical changes of TCB would be expected to occur once a GDD threshold was reached and there would be a small window for error. The studies at these two locations suggest that TCB first flight is also dependent on other currently unknown factors that need to be met before TCB can emerge.

Just as with first flight, there was a wide range in both the Julian day and accumulated GDD of TCB peak abundance for both locations. Peaks occurred as early as mid to late April and as late as early June for both locations. This wide range of dates could be caused by low count data, so GAM models were used to see if it could more accurately predict when peak abundance would occur by smoothing out the count data curves. GAM models, although they could typically predict peak abundance for a given year, they could not be consistently applied to predict future peak abundance or predict peaks abundance at other locations. Based on the work of Edwards et al. (2023), there was some expectation that—based on the small number of observations—anchor zeros would smooth out GAM models and make them more reliable at predicting the peak abundance of TCB flight. However, it became clear that anchor zeros at the end of a given year skewed the predicted peak abundance farther to the right than was justified.

This, in turn, caused peak abundance to be predicted later in the year than expected based on the observations. Anchor zeros may not have been the best choice for this data set as WDFW took recordings until they reached a near zero observations. Since a zero or near zero observation was not always captured at the beginning of each year's observation and the GDD growth rate is slower at the beginning of April/May than it is in June, perhaps having just the initial anchor zero in years where it was not captured would have smoothed the GAM model out better.

If more time and resources are available to make TCB observations in the future, it is recommended that observers go out more frequently starting in early to mid-April so that the likelihood of capturing first flight is increased and then reducing the visits to the standard two to three times per week and following current methods afterwards. Additionally, more information, such as local temperatures, should be recorded. It would be ideal to have daily temperatures recorded year-round at each habitat to more accurately determine the microclimate differences in GDD, thus potentially making GDD a more reliable predictor in the future. GDD is often used to monitor other species of lepidoptera, however there may be other factors playing a role in emergence (Chowdhury et al., 2021). Just as spongy moth rely on oak growth, TCB may rely on plant phenology to determine when they emerge (Foss & Rieske, 2003). It would be interesting to see if this was true as the reliance of their historic host plants, such as paintbrush species, have either been greatly diminished or have gone locally extinct in the fields where TCB currently reside (Grosboll, 2011).

It is important to reiterate that monitoring is important to this endangered species and the risk of extinction as climate change continues is high. Knowing what drives the life cycle of the TCB can better inform habitat restoration projects. If the timing of adult emergence and host plants productivity no longer align, then restoration projects may need to shift their focus such as they have for the planting of plantain. GDD has worked well for predicting many insect species emergence, including other lepidoptera species. Since this limited data cannot find an accurate model to predict emergence, captive populations should be monitored more closely to find a more exact GDD by using temperature data recorded in the greenhouses where TCB are reared and holding onto the pupa until they become adults. The life stage of their host greenhouse plants should also be documented to see if they play a role in emergence. Additionally other factors, such as precipitation, should be analyzed to see if any combination effects emergence as temperature alone does not.

TCB, like many butterfly species, are at risk of going extinct with the rise of climate change. In order to save these species it is important for conservation efforts to know how changing temperatures and habitat composition will affect their populations. Although temperature could not be directly linked to adult TCB emergence with this study, it is highly suspected that temperature, in concert with other factors are important consideration for TCB conservation and should be analyzed further.

Appendix I: JBLM R50 Julian Day GAM Predictions for Peak TCB Abundance

Below is list of GAM predictions using Julian day and various techniques on JBLM R50 site from 2010-2019. These techniques include a gamma = , gamma = 1.4, and a model using data with anchor zeros and gamma = 1. For each GAM, a predicted peak abundance (Pred.), a predicted minimum (Min) and maximum (Max) Julian day, p-value, R-squared Adjusted (R-sq Adj), the number of data points (n), and k values (n/2) were recorded. The anchor zeros add two to the n value and the k value is rounded up to the nearest whole number. All Julian day predictions were rounded up to the nearest whole number so there were no partial days.

	Julian Day Gamma = 1.4							**Julian Day Anchor 0 and Gamma = 1**						
Year	Min	Pred.	Max	p-value	R-sq Adj	n	K	Min	Pred.	Max	p-value	R-sq Adj	N	k
2010	125	129	132	0.359	0.358	6	3	128	130	135	0.252	0.344	8	4
2011	130	130	133	0.402	-0.028	7	4	132	140	147	0.505	0.096	9	5
2012	128	131	134	0.047	0.649	9	5	128	130	132	0.015	0.782	11	6
2013	123	126	129	0.195	0.444	9	5	125	126	127	0.006	0.873	11	6
2014	103	103	107	0.563	-0.098	8	4	110	118	126	0.62	0.016	10	5
2015	112	115	117	0.014	0.82	9	5	110	114	118	0.134	0.473	11	6
2016	109	112	115	0.143	0.476	9	5	106	111	115	0.315	0.243	11	6
2017	124	124	127	0.462	-0.079	5	3	128	133	140	0.328	0.308	7	4
2018	121	124	126	0.059	0.672	9	5	123	125	126	0.023	0.788	11	6
2019	117	120	123	0.046	0.764	7	4	120	121	123	0.002	0.948	9	5

	Julian Day Gamma = 1						
Year	Min	Pred.	Max	p-value	R-sq Adj	n	K
2010	125	128	131	0.231	0.448	6	3
2011	138	140	143	0.243	0.527	7	4
2012	128	130	133	0.043	0.696	9	5
2013	123	125	128	0.078	0.624	9	5
2014	114	120	125	0.565	0.049	8	4
2015	112	115	117	0.01	0.846	9	5
2016	109	111	114	0.09	0.592	9	5
2017	132	137	141	0.395	0.353	5	3
2018	121	123	125	0.024	0.778	9	5
2019	117	120	123	0.064	0.781	7	4

Appendix II: JBLM R50 GDD GAM Predictions for Peak TCB Abundance

Below is list of GAM predictions using GDD and varying techniques on the JBLM R50 location from 2010-2019. These techniques include a combinations of using gamma = 1 or gamma = 1.4 and models using data with and without anchor zeros. For each GAM, a predicted peak abundance (Pred.), a predicted minimum (Min) and maximum (Max) GDD, p-value, R-squared Adjusted (R-sq Adj), the number of data points (n), and k values (n/2) were recorded. The anchor zeros add two to the n value and the k value is rounded up to the nearest whole day. All GDD predictions were rounded up to the nearest whole number so there were no partial days.

	GDD Gamma = 1.4							**GDD Anchor 0 and Gamma = 1.4**						
Year	Min	Pred.	Max	p-value	R-sq Adj	n	k	Min	Pred.	Max	p-value	R-sq Adj	N	k
2010	152	159	167	0.037	0.83	6	3	148	163	181	0.206	0.383	8	4
2011	75	75	82	0.395	-0.023	7	4	40	40	55	0.447	-0.046	9	5
2012	126	137	149	0.131	0.513	9	5	123	137	151	0.166	0.419	11	6
2013	125	139	155	0.056	0.635	9	5	133	143	152	0.044	0.676	11	6
2014	93	93	102	0.329	0.018	8	4	48	48	65	0.477	-0.052	10	5
2015	149	149	156	0.15	0.168	9	5	174	189	200	0.153	0.441	11	6
2016	170	187	204	0.211	0.356	9	5	170	185	204	0.228	0.329	11	6
2017	116	116	126	0.341	0.064	5	3	133	160	198	0.435	0.193	7	4
2018	125	142	161	0.29	0.346	9	5	129	144	158	0.12	0.499	11	6
2019	146	157	169	0.024	0.842	7	4	155	164	176	0.032	0.805	9	5
	GDD Gamma = 1							**GDD Anchor 0 and Gamma =1**						
Year	Min	Pred.	Max	p-value	R-sq Adj	n	k	Min	Pred.	Max	p-value	R-sq Adj	N	k
2010	153	159	167	0.032	0.836	6	3	146	163	183	0.241	0.349	8	4
2011	75	75	82	0.395	-0.023	7	4	40	40	55	0.447	-0.046	9	5
2012	126	135	144	0.074	0.622	9	5	108	134	159	0.436	0.148	11	6
2013	125	139	152	0.045	0.703	9	5	46	46	63	0.473	-0.046	11	6
2014	93	93	102	0.329	0.018	8	4	48	48	65	0.477	-0.052	10	5
2015	180	187	194	0.025	0.779	9	5	103	103	118	0.311	0.015	11	6
2016	170	178	186	0.029	0.812	9	5	142	176	207	0.415	0.146	11	6
2017	131	158	180	0.397	0.275	5	3	60	60	81	0.461	-0.064	7	4
2018	123	131	142	0.039	0.781	9	5	51	51	68.7	0.459	-0.042	11	6
2019	144	153	163	0.023	0.89	7	4	137	164	194	0.54	0.114	9	5

Appendix III: Scatter Creek Wildlife Area GAM predictions of TCB peak abundance by Julian Day and GDD

Below is a table of TCB predicted peak abundance for both Julian day and GDD using gamma = 1 on the SCWA location from 2010-2023. For each GAM, a predicted peak abundance (Pred.), a predicted minimum (Min) and maximum (Max) peak abundance date, p-value, R-squared Adjusted (R-sq Adj), the number of data points (n), and k values (n/2 rounded up to the nearest whole number) were recorded. All predictions were rounded up to the nearest whole number to avoid partial days. Due to Covid-19 restrictions, not enough data was collected in 2020 to run GAM models.

	Julian Day Gamma = 1							**GDD Gamma = 1**						
Year	Min	Pred	Max	p-value	R-sq Adj	N	K	Min	Pred.	Max	p-value	R-sq Adj	n	k
2010	119	125	130	0.153	0.569	6	3	135	150	163	0.109	0.649	6	3
2011	148	152	156	0.046	0.421	16	8	138	151	163	0.03	0.464	16	8
2012	149	154	154	0.445	0.031	11	6	184	204	224	0.412	0.095	11	6
2013	122	124	126	0.002	0.94	8	4	126	137	148	0.02	0.825	8	4
2014	121	125	128	0.134	0.505	9	5	144	156	168	0.079	0.625	9	5
2015	111	115	119	0.104	0.463	11	6	198	208	221	0.028	0.672	11	6
2016	113	117	120	0.128	0.423	11	6	205	218	232	0.044	0.596	11	6
2017	135	138	141	0.063	0.782	7	4	147	163	180	0.05	0.812	7	4
2018	126	129	132	0.2	0.328	10	5	164	175	186	0.003	0.942	9	5
2019	114	114	116	0.2	0.133	8	4	138	138	146	0.037	0.539	7	4
2020	-	-	-	-	-	3	-	-	-	-	-	-	3	-
2021	116	118	121	0.025	0.884	7	4	129	135	142	0.003	0.974	7	4
2022	126	131	136	0.455	0.118	9	5	116	127	140	0.423	0.158	9	5
2023	128	129	131	0.121	0.624	6	3	123	134	145	0.063	0.749	6	3

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