Doctoral (PhD) dissertation

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Gödöllő, Hungary

2025

HUNGARIAN UNIVERSITY OF AGRICULTURE AND LIFE SCIENCES

DOCTORAL SCHOOL OF BIOLOGICAL SCIENCES

DEMOGRAPHIC ANALYSIS OF A BUTTERFLY POPULATION

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2025

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CONTENTS

LIST OF ABBREVIATIONS AND TERMS

age	number of days elapsed from the first capture of an individual (a minimum estimate for the real age)
AIC	Akaike Information Criterion
ĉ *or* VIF	Variance Inflation Factor
CI	confidence interval
CJS	Cormack-Jolly-Seber model
CO_2	carbon-dioxide
DOF	day of flight period, days elapsed since the first observed Clouded Apollo
GOF	Goodness of fit
initial (body) mass	the first measurement of the body mass of Clouded Apollos
initial thorax (width)	the first measurement of the thorax width of Clouded Apollos
LR	likelihood ratio test
(body) mass	body mass of Clouded Apollos (remeasured multiple times during butterflies' lifetime)
MRR	Mark-Release-Recapture
nobs_lev	levels of observers, level 1 = 1, level 2 = 2 or 3, level 3 > 3 observers were in the field
T_PREV_DAY	mean daily temperature of the previous day (between 9 am and 6 pm) of recapture, measured in the shaded areas of the study site
RH_PREV_DAY	mean daily relative humidity of the previous day (between 9 am and 6 pm) of recapture, measured in the shaded areas of the study site

T_DAY	mean daily temperature of the day (between 9 am and 6 pm) of recapture, measured in the shaded areas of the study site
RH_DAY	mean daily relative humidity of the day (between 9 am and 6 pm) of recapture, measured in the shaded areas of the study site
p	recapture probability
Phi or *Φ*	apparent survival
PIM	Parameter Index Matrix
probo	proboscis length of Clouded Apollos (measured only once during butterflies' lifetime)
rain_24H_max	total rainfall in 24 hours
RH_shadow_day_med	median daily relative humidity (between 9 am and 6 pm) measured in the shaded areas of the study site
RH_shadow_night_med	median night relative humidity (between 6 pm and 9 am) measured in the shaded areas of the study site
SE	standard error
SOL_RAD_sun_med	median daily solar radiation (between 9 am and 6 pm) measured in the open area of the study site
TEMP_shadow_day_med	median daily temperature (between 9 am and 6 pm) measured in the shaded areas of the study site
TEMP_shadow_night_med	median night temperature (between 6 pm and 9 am) measured in the shaded areas of the study site
thorax (width)	thorax width of Clouded Apollos (measured multiple times during butterflies' lifetime)
time	factorial covariate, 'full' CJS model where apparent survival and/or recapture probability were estimated for each sampling interval/occasion

VIF *or* ĉ	Variance Inflation Factor
wing	wing length of Clouded Apollos (measured only once during butterflies’ lifetime)
χ2	chi square

1. INTRODUCTION

In recent decades, the combined effects of global climate change (e.g. increasing mean temperature) and intensified human land use (e.g. habitat ploughing, urbanization, aggravating logging of trees or pesticide use) have led to the loss and deterioration of natural habitats worldwide (He et al., 2019; Mantyka-Pringle et al., 2011). To stop biodiversity decline, the most pressing questions today are if and how organisms are able to adapt to these rapid environmental changes and how population viability and individual fitness are affected by these changes. To answer these questions, it is essential to understand the life history of species and to identify the environmental factors that influence life history traits.

Demographic studies can analyse various life history traits of a species, including the number of offspring, age and size at maturity, age-specific mortality (senescence), and investment in reproduction or adult lifespan. These demographic parameters determine the population structure and dynamics, which may be influenced by environmental factors (temperature, humidity, solar radiation etc.), the availability of resources (food resources, shelter, etc.) and interactions between conspecifics and other species (Braendle et al., 2011; Stearns, 1992). Life history traits are related to each other through different trade-offs, i.e. survival and current reproduction, current or future reproduction or the number, size and sex of the offspring. These traits influence an organism's ability to survive (viability), to reproduce (reproductive success), and, ultimately, contribute to the overall measure of its evolutionary success, i.e. fitness. Reproductive success and survival are considered as major fitness components and they can be estimated by measuring life history traits (Braendle et al., 2011; Stearns, 1992). Reproductive success (total number of offspring a female produces in her lifetime) is critical for the survival of a population, but we can only successfully estimate this for some species.

In insects, the weight or number of eggs in females at eclosion or the total number of eggs laid by females are often used as reproductive success estimates (e.g. Campbell, 1962; Proshold, 1996). For butterflies, reproductive success is difficult, often impossible to measure in natural populations – indeed, we could not find such studies so far. However, studying apparent survival with mark–recapture method and assessing ageing in natural butterfly populations can be easier to implement (e.g. Carroll and Sherratt, 2017; Osváth-Ferencz *et al.*, 2017; Sielezniew, Kostro-Ambroziak and Kőrösi, 2020).

Survival probability of different species depends on a complex interplay of numerous factors of their ecosystem, key elements include both intrinsic and extrinsic factors (Baudisch, 2008; Stearns et al., 2000; Williams & Day, 2003). Intrinsic factors include genetic adaptability, species-specific traits, and individual health, influencing an animal's ability to overcome challenges (Kirkwood & Austad, 2000). Extrinsic factors, often more variable, include environmental changes, habitat quality, predation pressures, infections, resource availability, and human interference (Kirkwood & Austad, 2000). Climate change, habitat loss, pollution, and habitat deterioration exacerbate threats to survival. Moreover, social dynamics within a species, such as cooperation or competition, also impact an animal's odds of survival (Silk et al., 2003; Vickruck & Richards, 2019). The interplay among these multifaceted factors shapes an animal's resilience and survival prospects within its environment.

Senescence, the deterioration of biological functions with age, a crucial mechanism of decreasing fertility and survival probability through an individual's life, has been a subject of interest in understanding the life history of insects. In the wild, the demonstration of senescence in insects has been challenging due to their short lifespans and high mortality rates, which limit the evidence for senescence (Nussey et al., 2013; Zajitschek et al., 2020). Recent results highlighted the need for longitudinal studies to comprehend the causes and fitness consequences of life history variation in wild insects in order to study trade-offs between body size, reproductive effort, and senescence rates (Bonduriansky & Brassil, 2005; Rodríguez-Muñoz, Boonekamp, Liu, Skicko, Fisher, et al., 2019). We distinguish three types of senescence: actuarial senescence (decreasing survival rate), reproductive senescence (reduction of reproductive success) and phenotypic senescence (phenotypic recession) (Nussey et al., 2013). As we mentioned above, reproductive success cannot be estimated easily/properly in natural butterfly populations, but actuarial and phenotypical senescence estimates are feasible.

To estimate actuarial senescence, mark-recapture studies were usually conducted in natural populations of different species, e.g. ungulates (e.g. Gamelon *et al.*, 2014), birds (e.g. Barbraud, 2019), amphibians (e.g. Cayuela et al., 2020, 2019), or butterflies (e.g. Sielezniew et al., 2020). Phenotypic senescence has been observed in natural populations of a wide range of vertebrates (for a review of mammals and birds see: Nussey *et al.*, 2013), however, our knowledge is scarce about senescence and lifespan in natural populations of invertebrates, which are mostly studied in the laboratory (Nussey et al., 2013; Zajitschek & Bonduriansky, 2014). Different body size parameters (i.e. body mass, thorax width, wing length), may serve as suitable proxies to assess body condition of butterflies, from which phenotypic senescence can be estimated. Stjerholm and colleagues detected decreasing thorax mass with increasing wing wear (estimation for individual's age) in individuals captured from natural populations, but without repeated measurements of body size (Stjernholm et al., 2005). Karlsson observed decreasing abdomen mass for both sexes and thorax mass for only females with increasing wing wear in *Pararge aegeria* and *Speyeria mormonia* butterflies, and increasing body and thorax mass with increasing wing wear for both sexes in *Heliconius hecale* butterflies (Karlsson, 1994) in individuals captured from natural populations and also without repeated measurements of body sizes. To detect phenotypic senescence, repeated measurements of different body variables along individuals' lifespan is necessary. Boggs and colleagues detected body mass change with age in laboratory populations of butterflies (Boggs, 1997), and it seems that a relationship between age and thorax mass also exists (Karlsson, 1994; Stjernholm et al., 2005), therefore these body sizes may be promising variables for detecting senescence. Wing length and wingspan, both strongly correlated with body size, are widely used proxies for flight capacity in butterflies (Sekar, 2012), and they can also indicate individual fitness (Kingsolver, 1999).

In demographic studies, it is important that the species under study can be easily observed. Lepidoptera have become model organisms in ecological and evolutionary research in recent decades (Boggs et al., 2003), and are among the most intensively studied groups of insects (Insecta), which make up the bulk of terrestrial biodiversity. Most butterfly species have been recognized as valuable indicators of environmental changes due to their rapid response to environmental variations (da Rocha et al., 2010; Rákosy & Schmitt, 2011; Thomas, 2005) and are ecologically important due to their high diversity (da Rocha et al., 2010; van Swaay et al., 2006).

Demographic studies on insects, often conducted in laboratory circumstances for short (1–3 years) periods, pose challenges when projecting findings to wild populations. Environmental factors can influence fitness, i.e. resource availability affecting body size and ageing or weather variables affecting ageing. Genetic variations and environmental factors between laboratory and wild populations may impact senescence rates (Finch & Kirkwood, 2000; Kawasaki et al., 2008; Kirkwood & Austad, 2000; Matos & Avelar, 2001). Short-term studies do not allow to separate within-individual ageing patterns from between-individual heterogeneity (Van de Pol & Verhulst, 2006), therefore, long-term studies in natural populations are needed to correctly assess these life history traits in insects.

Understanding the impact of environmental factors on life history traits and demographic parameters is important in predicting the future distribution and the viability of populations or species (Settele et al., 2009; WallisDeVries et al., 2011).

2. OBJECTIVES TO ACHIEVE

My research focuses on the demography analysis of a natural population of Clouded Apollo butterflies (*Parnassius mnemosyne*, Linnaeus 1758, Lepidoptera: Papilionidae), which was studied using mark-recapture and *in situ* non-invasive body size measurements in the field. The univoltine Clouded Apollos are relatively easy to observe, capture and mark, and they are robust enough for *in situ* non-invasive measurements of body sizes without apparent harm.

The main aims of my doctoral research were to:

1) investigate the change of body mass and thorax width with age in a natural population of the Clouded Apollo butterfly over seven consecutive years,
2) investigate the relationships between wing length and other body sizes,
3) uncover the relationships between survival probability and age, day of the flight period, weather variables and body sizes,
4) uncover the relationships between recapture probability and age, day of the flight period, weather variables and body sizes.

I was also seeking answers to the following questions:

- Did body mass and thorax width decline with age (phenotypic senescence)? If so, was this decline linear or polynomial?
- Were there differences between the sexes in initial body sizes and how did body sizes change with age?
- Was the date of first capture of an individual related to its body size and the rate of senescence?
- Was wing length correlated with other body size variables and phenotypic senescence?
- How did survival and recapture probability change with age and the progress of the flight period?

- How were survival and recapture probability related to daily mean temperature and relative humidity?
- How were survival and recapture probability related to initial body mass, initial thorax width, wing length and proboscis length of individual butterflies?

3. LITERATURE OVERVIEW

The main aim of demographic studies is to explore the structure (number of individuals, density, spatial distribution, age distribution, etc.) and dynamics (fertility, mortality, growth rate, etc.) of populations and to understand the factors that influence them. Variables of population dynamics determine population structure, since the number of individuals depends essentially on birth rate, survival, immigration and emigration (Stearns, 1992).

3.1 Life history

Organisms employ a wide variety of strategies and adaptations in their life histories to navigate their existence from birth to death. These life histories are shaped by the complex interaction of evolutionary forces and environmental pressures, resulting in a spectrum of reproductive, developmental, and survival tactics across species. Key components include the timing of reproduction, the number and size of offspring produced, parental investment, age at maturity, and lifespan.

Life history traits are limited by intrinsic factors, like trade-offs and constraints. Trade-offs involve the allocation of resources among competing functions, leading to fitness benefits in one trait at the expense of fitness costs in another. Constraints, on the other hand, are described as absolute limits or biases on trait expression and combination, and are often the results of physical factors, developmental properties, or unforeseen events. The distinction between trade-offs and constraints is not strict, with trade-offs often considered as one type of constraint. In the context of life history, constraints typically refer to phylogenetic, lineage-specific characteristics that impose absolute limits on trait expression within a particular organismal group (Braendle et al., 2011).

For instance, r-selected species often prioritize high reproductive output with numerous, smaller offspring and minimal parental care, suited for unstable or unpredictable environments. Conversely, K-selected species invest heavily in few, larger offspring with extensive parental care, thriving in more stable and predictable habitats (Stearns, 1992). Life history strategies are also influenced by ecological factors such as resource availability, predation pressures, and environmental stability. Understanding these diverse life history strategies provides critical insights into the evolutionary trade-offs that shape the survival and reproductive success of animals within their ecological niches (Cohen et al., 2017).

3.2 Life history of insects

There are often trade-offs between current reproduction and future survival or between the quantity and quality of offspring (Stearns, 1989), which is described by the allocation of resources towards reproduction at the expense of survival, or vice versa, as insects face ecological challenges and environmental constraints (Boggs & Freeman, 2005; Jervis et al., 2007; Stjernholm et al., 2005). The trade-off between reproduction and survival is further influenced by factors such as egg maturation, flight endurance, dispersal, and life history variation, shaping the evolutionary outcomes and ecological dynamics of butterfly populations (e.g. Jervis, Boggs & Ferns, 2007; Berger, Walters & Gotthard, 2008; Karlsson & Johansson, 2008). However, the physiological and energetic costs associated with reproduction and flight performance highlight that during trade-off, butterflies must navigate to optimize their life history strategies (e.g. Hughes, Hill & Dytham, 2003; Hanski, Saastamoinen & Ovaskainen, 2005). Understanding the relationship between fitness (reproductive success and survival), and individual quality and environmental influences is crucial for comprehending population resilience, specifically its viability.

Reproductive success in insects can be studied by measuring the number of laid eggs (fecundity), survival rate of eggs and larvae or mating success (e.g. Campbell, 1962; Proshold, 1996). These are difficult to measure because of the small size of larvae and eggs, which makes them hardly visible for the human eye, especially in natural circumstances, and because females often move to a hidden place to lay eggs, so fecundity – estimated with the number and total weight of eggs laid – is measured mostly in laboratory conditions (e.g. Proshold, 1996). Moreover, their small body size and either cryptic or, on the contrary, highly active lifestyle often make it challenging to observe their mating behaviour and accurately estimate mating success. In butterfly populations, fecundity is difficult to measure in natural conditions, so the viability of the population is studied through the survival of adults. Survival as a precondition for reproduction is one of the key life history components and depends on the life history strategies of the organisms, which may respond to environmental changes (e.g. Karl & Fischer, 2009).

Survival and reproductive success of butterflies are intricately linked to their fertility and the timing of egg-laying events. The ability of butterflies to survive until the next egg-laying event is crucial for achieving higher reproductive success. The way of egg-laying in butterflies, i.e. egg-clustering or single-laying, can also affect the fecundity and survival probability of adults and offspring. Certain butterflies attach single eggs to the tip of the leaves or grass, or other plant parts (e.g. *P. mnemosyne*, Hungary, Gór et al., 2023a or *Melanargia occitanica*, García-Barros & Fartmann, 2009) or drop them one by one to the ground (e.g. *P. mnemosyne*, Sweden, Wiklund, 1984 or *Melanargia galathea*, García-Barros & Fartmann, 2009). Other butterflies lay their eggs in clusters of fewer than ten eggs (e.g. *Heteropterus* spp., *Archon* spp. or *Boloria eunomia*), or in piles underside of leaves (e.g. *Araschnia levana*), or both small batches and single eggs (e.g. *Zerynthia polyxena* or some *Lycaena* species), or in larger batches (e.g. *Euphydryas* spp. or *Nymphalis polychloros*, García-Barros & Fartmann, 2009). All strategies have their advantages and disadvantages. Single-laying species’ lifespan of females can affect how many eggs females can lay during a flight season (Doak et al., 2006; Kőrösi et al., 2008). In case of batch-laying species, the size of clusters may be species-specific (García-Barros & Fartmann, 2009), or depend on individual variance, i.e. number of the eggs may depend on the resources or state of females (Wahlberg, 1995; Wahlberg et al., 2004) or weather during oviposition (Courtney & Duggan, 1983). Large egg-clusters could be advantageous for offspring, because the inner eggs are protected against parasitoids or diseases (Friedlander, 1985) and desiccation is slower than in smaller egg batches (Clark & Faeth, 1998), or could be disadvantageous because of the shared food resources or cannibalism (Courtney & Duggan, 1983).

3.3 Senescence in insects

Senescence, the degradation of physiological function and the decline of fitness with age (Rose, 1994) is found in all living organisms. Environmental stressors, such as extreme heat, may accelerate ageing, thus impacting demography, and ultimately, the viability of populations (Brunet-Rossinni & Austad, 2005). Rapid human-induced changes in natural habitats impose evolutionarily new stressors and this may have detrimental consequences to organisms, and accelerated ageing due to stressors may be an important factor in population decline.

We distinguish actuarial senescence (decreasing survival rate), reproductive senescence (reduction of reproductive success) and phenotypic senescence (phenotypic recession) (Nussey et al., 2013). Senescence in vertebrates is widely studied, as extensively reviewed in Nussey *et al.* 2013, but we have only scarce information about natural invertebrate populations, which have primarily been studied in laboratory settings (Nussey et al., 2013; Zajitschek & Bonduriansky, 2014). Because of the differences in physiology, reproduction, selection on late life performance (Zajitschek et al., 2020) or trade-offs in senescence and other life history traits (Stearns, 1992) in different taxa, we cannot draw general conclusions about senescence.

Actuarial and reproductive senescence in a wild insect population was demonstrated for the first time relatively recently in Antler Flies (*Protopiophila litigate*, Bonduriansky and Brassil, 2002). Since then, further studies have revealed age-related increase in mortality rates in wild insect populations, but there was no general pattern in senescence. In Honey Bees (*Apis mellifera*) forager mortality increased with age (Dukas, 2008). Antler Flies experienced rapid and costly ageing in the wild (Bonduriansky & Brassil, 2002), and large male body size was associated with high mating rates in early life and with a heightened rate of reproductive ageing (Bonduriansky & Brassil, 2005). In laboratory circumstances, Antler Fly males mated more often, lived longer, but aged faster than in wild, genetically similar populations (Mautz et al., 2019). In a natural population of Field Crickets (*Gryllus campestris*) both actuarial and phenotypic senescence were observed (Rodríguez-Muñoz et al., 2019). Azure Damselfly (*Coenagrion puella*) mortality rates increased with age in a natural environment (Sherratt et al., 2010), and in other odonate species actuarial senescence was also observed (Sherratt et al., 2011). *Anopheles gambiae* mosquitoes' mortality increased with age faster in wild/semi-wild populations than in protected laboratory conditions (Ryan et al., 2015). In Neriid Flies (*Telostylinus angusticollis*), faster actuarial ageing and shorter lifespan were revealed in wild males compared to genetically similar males reared in laboratory populations (Kawasaki et al., 2008). In 22 butterfly species, actuarial ageing was detected in captive and in natural populations (Carroll & Sherratt, 2017), furthermore actuarial senescence was also observed in natural populations of the Large Blue butterfly (*Maculinea arion*, Osváth-Ferencz et al., 2017) and in Meleager's Blue butterflies (*Polyommatus daphnis*, Sielezniew et al., 2020).

Phenotypic senescence, the degradation in phenotypic traits with age, may also imply fitness costs through declining performance (Stearns, 1992). To study phenotypic senescence in natural populations, repeated measurements of phenotypic traits throughout an individual's life are required. This is challenging in insects due to their small and fragile body, high mobility and cryptic life cycle (Nussey et al., 2013; Zajitschek et al., 2020). The only study we found is Rodríguez-Muñoz *et al.*, 2019, on the singing activity of male Field Crickets that declined with age under natural conditions in five of nine study years. We only found one study that demonstrated a decline in body mass and reproductive effort of three butterfly species in laboratory conditions (Boggs, 1997).

3.4 Relationships between body size and life history traits in insects

Survival and rate of senescence in insects can be affected by developmental and adult parameters (egg size, adult body size etc.) (Boggs, 2009). Body size is partly genetically determined and partly influenced by environmental factors in insects (Nijhout et al., 2014). Currently, there are no established standardised measurements of body sizes that serve as predictive indicators of ageing; therefore, the identification of such parameters necessitates comprehensive measurement across multiple variables.

Longer lifespan may be associated with larger body size (e.g. in geometrid moths, Holm *et al.*, 2016) and environmental conditions may affect the relationship between body sizes and lifespan (Norry & Loeschcke, 2002). Larger body size may also mean increased fecundity in insects (Karlsson & Wickman, 1990; see Honěk, 1993 for a review) and it was also found in some butterfly species (Bauerfeind & Fischer, 2008; Oberhauser, 1997). Body size also might influence survival rates: a higher amount of resources accumulated as body reserves can be later used for resource allocation (Boggs, 1981). Resource re-allocation can cause a reduction of body size in butterflies (Boggs, 1981), e.g. decline of thoracic mass can be caused by nitrogen allocation from thoracic muscles for reproduction or for somatic maintenance (Karlsson, 1994, 1998; Stjernholm et al., 2005). In some butterfly species, an age-dependent decrease in nitrogen content of the thorax was revealed (Karlsson, 1994; Stjernholm & Karlsson, 2000) and females have been proven to transfer resources from thorax into eggs (e.g. *Pieris napi*, Karlsson, 1998, 1994). As deterioration of flight muscles may affect flight performance (Ahman & Karlsson, 2009), a decrease in thorax mass with age can be regarded as phenotypic senescence. Decrease in body mass in males can also be caused by transferring spermatophores and sometimes nuptial gifts during mating to females, or in females when laying eggs.

Males are likely to allocate more resources in reproduction than in survival and therefore age faster and live shorter than females (Sielezniew et al., 2020). Longer lifespans in females may allow more eggs to be laid, indirectly increasing fecundity (Bauerfeind & Fischer, 2008; Kőrösi et al., 2008; Rolff, 2002). Furthermore, as in anisogamous species, in the Meleager's Blue butterflies, the females' interest is to mate with the best possible male once they have invested a lot of resource to the eggs, while the males invest less in sperms, so their aim is to mate with as many females as possible in the shortest possible time (Sielezniew et al., 2020).

In butterflies, the quantity of larval food can also affect the allocation of resources between body parts, with females allocating more resources to the abdomen if the larva can accumulate more nutrients, thus increasing reproductive potential (Karlsson & Wickman, 1990), and males to the thorax and wing, thus increasing their flight and manoeuvring abilities, which are important in finding mates and escape predators (Chai & Srygley, 1990). Absolute thorax mass decreases with age according to several studies (Karlsson, 1994; Stjernholm et al., 2005), i.e. the relationship between body size and survival can be well examined by thorax mass. Both thorax and abdomen mass decrease with increasing wing wear (estimations for individuals' age). Females use the nitrogen content of the thorax for egg maturation, and thus the thorax decreases faster than in males (Karlsson, 1994). Phenotypic differences also affect fecundity and reproductive success through survival, with important relationships between body size and survival probability of offspring in insects, and between larval mass and fecundity, where a linear relationship has been observed (Tammaru et al., 2002).

The above- mentioned studies were conducted in laboratory conditions during a relatively short time period, which may be not appropriate to draw conclusions to wild populations, because circumstances, i.e. weather variables or resource availability may all affect body size and senescence, and might cause interannual variation (Evans, 2000). To our best knowledge, there are no widely accepted body size measurement protocols for live butterflies to date and we could not find any long-term studies with repeated measurements of phenotypic traits in wild insects.

3.5 Relationship between weather and life history traits in insects

Global climate change is one of the most serious problems in the last two centuries humanity is facing. Human activities have driven global temperatures 1.26 °C above pre-industrial levels (Betts et al., 2023), faster than at any time in the last 2000 years. Without taking rapid action to mitigate emissions, we risk exceeding the critical 1.5 °C limit within the next two decades. On current trends, temperatures could rise by up to 3 °C by the end of the century (IPCC, 2023).

The survival and life history of animals are convolutedly linked to environmental factors like different weather variables. Changing weather has been found to shape life history traits, population dynamics, and individual heterogeneity in different animal populations (Jenouvrier et al., 2015; Morris et al., 2008; Sorel et al., 2023).

Insects, as ectothermic species, are extremely threatened by increasing temperature, because many of them do not have enough physiological tolerance to survive continuous exposure to the current maximum temperatures experienced in their habitats (González-Tokman et al., 2020). Temperature influences insect physiology, behaviour, and fitness (Roux et al., 2010). It can affect herbivorous insects indirectly through their host plants (Bale et al., 2002). For example, butterflies reared on drought-stressed host plants showed reduced survival or reproductive success (Gibbs et al., 2012). Butterflies, being ectotherms, are good model organisms for studying the effects of weather variability (Wikstroem & Bergman, 2009), and their short generation time gives them a rapid response to changes in environmental conditions. Several studies have found that the lifespan of butterflies decreases with increasing temperature (Karl & Fischer, 2009; Karlsson & Wiklund, 2005; Klockmann et al., 2016). A possible reason for this could be that at higher temperatures the metabolic rate of ectotherms will be higher (e.g. Terblanche, Klok & Chown, 2005) which results in shorter lifespans via senescence (Melvin et al., 2007). It was also found that elevated temperature can cause reduced immune response in butterflies (Karl et al., 2011), while a recent study found that starvation resistance, and especially survival, declined with increasing temperature in the Monarch butterfly (*Danaus plexippus*, Ragonese et al., 2024).

From laboratory studies and short-term field observations, we know that the activity and distribution patterns of many butterfly species can be influenced by local weather (Merckx et al., 2006, 2008), with temperature and the number of hours of sunshine being the most important factors (Wickman, 2009). Butterflies show decreased activity below 19 °C, while at higher temperatures they are more active in less sunshine (Wikstroem & Bergman, 2009). The level of activity is strongly correlated with the intensity of solar radiation (Meyer & Helminger, 1994). In *Lycaena tityrus*, there was also variation between sexes, with females surviving longer at higher temperatures than males and generally better able to withstand extreme conditions (Karl & Fischer, 2009). In contrast, Vlašánek et al. found that survival increased for Clouded Apollos in warm, humid weather (Vlašánek et al., 2009). In *Parnassius clodius*, as the population aged towards the end of the flight season, survival probability decreased, with an increase in the proportion of females in the population (Auckland et al., 2004).

Relative humidity also plays a major role in the survival of insects, and previous studies have shown that insects, particularly Fig Wasps (*Ceratosolen* species), exposed to humid environments lived longer than those exposed to dry conditions (Gigante et al., 2021). It has been revealed recently, that flying honey bees avoid overheating by evaporative cooling and thus desiccation likely limits their foraging under hot and dry weather conditions (Glass et al., 2024). The fitness consequences of climate change for butterflies, including their ability to achieve body temperatures needed for flight, oviposition, and overall fitness, are influenced by their phenology, plasticity, and evolution (Kingsolver & Buckley, 2018; Nussey et al., 2013).

The effect of weather on butterfly activity and behaviour can be well studied using the mark-recapture method. Slamova and colleagues found that time spent basking decreased with increasing temperature, while time spent foraging increased, and male mating activity was reduced by high humidity (Slamova et al., 2011). Inactivity is observed at extreme temperatures, with animals retreating to the shade in hot weather (Casula & Nichols, 2003) and taking a long time to warm up again in severe cold (Vlašánek et al., 2018). The number of warmer days is also strongly correlated with the average length of stay in the habitat and the emigration rate, i.e. the year-to-year variation in dispersal rate depends on the weather during the flight season (Kuussaari et al., 2016).

3.6 Mark-release-recapture method

Mark-release-recapture (MRR) studies represent a key method in understanding butterflies' survival rates, population dynamics or dispersal patterns. In these studies, butterflies are captured, marked with a unique identifier (using tags or dyes), then released back into their natural habitat. Subsequent recaptures or resights allow researchers to estimate population size, movement, and survival rates over time.

MRR studies have contributed significantly to butterfly ecology and conservation efforts (Ehrlich et al., 1975; Ehrlich & Hanski, 2004), offering insights into population structure (e.g. Fischer, Plachter and Beinlich, 1999), population size (e.g. Verovnik *et al.*, 2013), abundance, activity (e.g. Casula & Nichols, 2003) or dispersal between habitat patches (e.g. Roland, Keyghobadi & Fownes, 2000; Auckland, Debinski & Clark, 2004; Kuussaari *et al.*, 2016). However, in most cases, only average lifespans are provided, estimated from survival (e.g. Bubová *et al.*, 2016), but this can be misleading, as they do not provide information on the distribution of lifespan, and individuals with longer lifespans than average are expected to produce more offspring.

These studies are well suited to observe both actuarial and phenotypic senescence, as the markings allow us to repeatedly measure different body sizes of animals and track changes in these body sizes. Moreover, from the recapture data, we can estimate the survival and recapture probability of the animals.

3.7 Studies in laboratory vs. in nature

Most of the above-mentioned studies of insects about life history and/or senescence were conducted in a laboratory and/or in a controlled environment and lasted only 1–3 years. Drawing conclusions from laboratory studies to wild populations could be problematic, because fitness components and resources that determine body sizes and senescence are all affected by the experienced environmental conditions which may cause interannual variation in insect size (e.g.

Alcock, 1984; Evans, 2000). This seasonal variation is mostly affected by developmental temperature (e.g. David *et al.*, 1997), resource availability (e.g. Gibbs et al., 2012) or stressful conditions such as desiccation (e.g. Lighton et al., 1994), intraspecific competition (e.g. Heinrich & Bartholomew, 1979) or predation intensity (e.g. Nylin & Gotthard, 1998). Furthermore, laboratory populations are often genetically different from their conspecifics in the field due to genetic drift or adaptation to laboratory conditions (Matos & Avelar, 2001), potentially resulting in differences in ageing (Kenyon, 2005; Kirkwood & Austad, 2000). Even genetically similar wild and captive groups of a population may show different degrees of senescence (Kawasaki et al., 2008). In addition, studies under controlled circumstances did not explain the variance in intra- and interspecific longevity and ageing, nor how environmental variance affects the rate of ageing (Flatt et al., 2013).

Senescence shows high variability among years in natural circumstances as individuals have to face different environments in each year. As senescence is a within-individual process, we shall conduct long-term studies (Rodríguez-Muñoz et al., 2019) to control between-individual heterogeneity which can mask within-individual senescence (Van de Pol & Verhulst, 2006).

Therefore, we think that it is important to study butterflies also in their natural habitats, as both their survival and reproductive performance can be affected by many variables, from weather factors to variation in body size to the quantity and quality of available resources (see also Boggs, 2009). Mark-recapture has been widely and successfully used in field studies to investigate populations of butterflies, but only few studies have investigated the temporal variation of demographic parameters (e.g. survival) and their dependence on weather and/or body size.

4. MATERIALS AND METHODS

4.1 Study species

The Clouded Apollo butterfly (*Parnassius mnemosyne*) is a widespread species in the Western Palearctic realm. Its polycentric distribution area ranges from the Pyrenees in Northern Spain to the Tien Shan Mountains, Kyrgyzstan (Weiss, 1999), and in the temperate zone of Asia, as far as Tajikistan and Northwest China (van Swaay et al., 2012). However, in the last century, its distribution areas have been constantly shifting further North (Parmesan et al., 1999). This butterfly usually occurs below 1500 m a.s.l., but in Central-Asian mountains it has also been observed at higher altitudes (van Swaay et al., 2012). In Central Europe, it inhabits woodland clearings and meadows, rich in flowering plants and open sunny areas, surrounded by woods (Weiss, 1999). This protandrous, univoltine species' flight period runs from late April to the beginning of June in Hungary (Gergely et al., 2018). Adult Clouded Apollos spend plenty of time on feeding (Szigeti et al., 2018a). Males search for females by patrolling, and during mating they may produce a large, presumably costly sphragis attached to the females' copulatory orifice to prevent re-mating (Gór, Fónagy, et al., 2023; Gór, Lang, et al., 2023; Vlašánek & Konvička, 2009). Mated females of the studied population lay eggs one-by-one on leaf litter or grass near patches of *Corydalis* species (Gór, Fónagy, et al., 2023). The eggs overwinter, and caterpillars hatch in early spring, feeding exclusively on various *Corydalis* species. During this period, weather conditions may affect their feeding success and adult body size (Bergström, 2005). After reaching the appropriate size, caterpillars pupate wrapped in leaves (van Swaay et al., 2012).

Sexual dimorphism is clearly visible in adults, females having yellow hair on the back of the head and yellow scales along their abdomen, and the dorsal thorax and abdomen are less hairy in females than in males (Figure 1).

Figure 1: Female (left) and male (right) Clouded Apollos (Photo: Tamás Nestor)

Most Clouded Apollo populations are declining, due to the global climate change and human land use intensification or abandonment of traditional management, which lead to the loss and deterioration of natural habitats. This species can be found on the IUCN Red List, in Hungary, it is protected by the law, and their habitats are protected by the Bern Convention (van Swaay et al., 2012).

4.2 Study site

Fieldwork was conducted between 2014 and 2020 at Hegyesd, a 0.5-hectare meadow (47°45'22.7"N, 19°2'53.4"E, at 295 m a.s.l.) in the Visegrádi-hegység, Central Hungary. Insect-pollinated flowering plants were heterogeneously distributed all over the meadow, surrounded by oak wood (*Quercus cerris*). There is a transition zone between forest and open grassland with specific microclimatic conditions and structural elements such as trees, leaf litter and dried grass, which are important for the development of the Clouded Apollos' caterpillars (Habel et al., 2022).

4.3 Sampling methods

4.3.1 Mark-release-recapture method

Mark-release-recapture (MRR) was used to sample the population. Fieldwork covered the whole flight period every year. Sampling started when the first Clouded Apollo adults appeared and lasted until the last individual was on wing. Data collection was conducted by 2–5 people every day during the flight period, as weather allowed. For the survey, observers followed the same routes which had been systematically distributed in the meadow to reduce trampling (Szigeti et al., 2016). Butterflies were captured with butterfly nets and were marked with a unique colour code and a number; the code was placed ventrally on the transparent tips of both forewings with Edding® paint markers, the number was written on the ventral side of both hindwings with Edding® permanent markers (Figure 2). Colour codes could be seen from both sides of the wing.

Figure 2: Male Clouded Apollo with markings at the apex of forewing (colour code) and on the hindwing (number) (Photo: Ádám Gór)

We surveyed the meadow several times a day and tried to catch all unmarked Clouded Apollos. As it is a small, closed population (Table 10), we assumed that butterflies were captured soon after their eclosion and their detectability did not vary among individuals and through time.

A preliminary analysis of the 2016–19 period's data on our study population using Jolly-Seber models estimated that ~90% of the individuals in this relatively closed population had been captured at least once in each year (author's unpublished data), suggesting that individuals present in the population were mostly detected.

4.3.2 Body size measurements

Initial body mass and initial thorax width are intended to represent the body size at hatching. In our research, the first measurement for most butterflies was taken within hours of hatching (as their yellow wing colour and/or soft wings are characteristic of the hours following emergence from the chrysalis). However, due to technical reasons, the initial measurement for some butterflies was taken a few days after hatching, at the first capture. Between 2014 and 2016, initial body mass measurements were performed every third day, not always at first capture. In further years, it was measured at first capture, along with thorax width and wing length. We believe that this does not significantly affect the study, as the objective was to examine a trend.

We attempted to recapture all marked individuals to repeat body mass and thorax width measurements every third day. We measured body mass with a Mettler-Toledo, NewClassic MF JS303G scale (Mettler-Toledo AG, Laboratory & Weighing Technologies, Switzerland) with 1 mg precision (Figure 3/a1, a2). Thorax width was measured twice at each occasion with callipers to 0.1 mm (Figure 3/b1, b2). Wing length was the average length of both forewings measured from base to the apex with a commercial plastic ruler (2014–15; resolution: 1 mm) or a printed ruler (2016–20; resolution: 0.2 mm) (Figure 3/c). Proboscis length was available in 2015–19, which we measured from photomacrographs (Szigeti et al., 2020) using the FIJI/ImageJ software (Schindelin et al., 2012). Alive, non-sedated butterflies were mounted on a board with clips and a scale, then we uncoiled the proboscis and extended it over the board with a hooked pin. At least two pictures for each individual were taken (Figure 3/d). If we measured a variable twice in a single measurement session, we used averages for all analyses. After marking and measuring, butterflies were instantly released. Clouded Apollo is a relatively robust species which allows repeated measurements of body sizes without apparent harm.

Handling and the associated measurements were done by János Kis, except for body mass which was measured by several people. All proboscis measurements from photographs were carried out by Flóra Vajna.

Field work was licensed by the Hungarian nature conservation authorities: KTVF: 31430/2014.

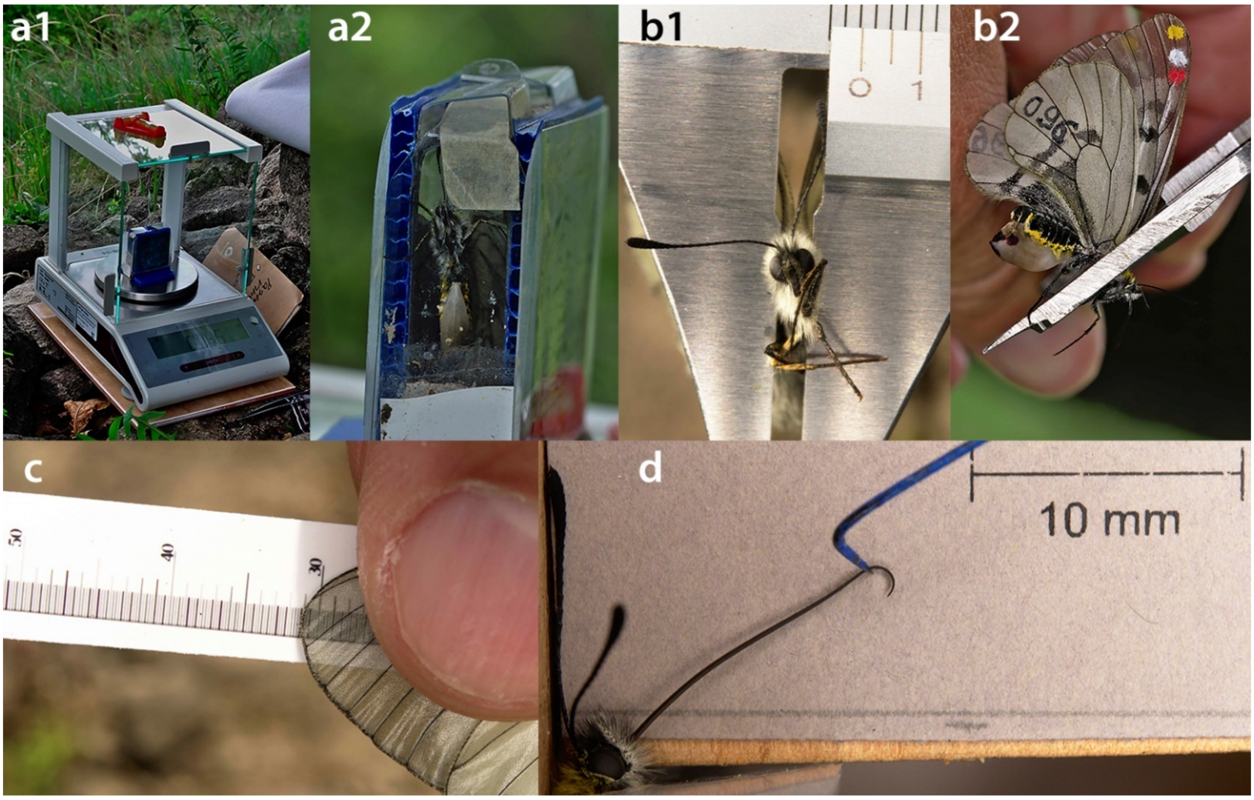


Figure 3: Body mass (a1, a2), thorax width (b1, b2), wing length (c) and proboscis length (d) measurements on the field (Photo: János Kis)

4.3.3 Weather data collection

Temperature and relative humidity were recorded every 10 minutes with a VOLTCRAFT DL-121TH multi-channel data logger (Figure 4/a) between 2014–2018, and with a VOLTCRAFT DL-210TH multi-channel data logger (Figure 4/b) since 2019, installed permanently in the shaded areas of the study site.

Figure 4: VOLTCRAFT DL-121TH (a)
and VOLTCRAFT DL-210TH (b) multi-channel data logger

4.4 Statistical analyses

All statistical analyses were done using ‘nlme’ (v3.1-152; (Pinheiro et al., 2021), ‘MuMIn’ (v1.43.17; Bartoń, 2020) and ‘RMark’ packages (Laake, 2013) in R 3.6.1 (R Core Team, 2021). The number of individuals was different for the Linear Mixed Effects model and for the Cormack-

Jolly-Seber model, due to the different conditions of entry to the models (see Section 4.4.1.1 and 4.4.2.1,Table 1Table 2).

4.4.1 Body size change analyses with age

4.4.1.1 Data preparation

We defined ‘age’ as the number of days elapsed from the first capture of an individual, which was a minimum estimate for the real age. Age^2 was used to model a non-linear relationship between age and body size. ‘Mean(age)’ and ‘mean(age^2)’ were the averages of all age and age^2, respectively, data at the time of measurements of an individual. These ‘mean variables’ enabled us to distinguish the within-subject and between-subject effects (Van de Pol & Verhulst, 2006; Van de Pol & Wright, 2009). Butterflies not only had different body sizes, but also their age at measurement showed high variation. By using ‘mean(age)’ and mean(age^2), we could test if body size declined with age (within-individual effect) and if individuals measured at an older age had smaller body size (between-individual effect). The variables mean(age) and mean(age^2) are the difference of within-individual effect, and between-individual effect of age (linear) and age^2 (quadratic), respectively.

The ‘first capture’ variable was the day of the flight period when an individual was captured for the first time, whereas ‘mean(first capture)’ was the annual average of the days of first captures. Distribution of ‘first capture’ was different among years and ‘mean(first capture)’ was correlated with the length of the flight period (see variation in flight period length in Table 10). By using ‘mean(first capture)’ as a covariate, we could also test if body size variation among years was related to the variation in flight period length.

Individuals with at least two measurements of body mass and thorax width were included in this model, therefore data on some of the butterflies in the entire sample had to be omitted.

4.4.1.2 Analyses

Data from the seven study years were pooled and analysed within one model. First, we built a linear mixed-effects model for each response variable (body mass and thorax width) including age, age^2, mean(age), mean(age^2), first capture, mean(first capture) and wing length, the interactions between these variables and sex. We also tested the interactions between wing length and age and between first capture and age. In the case of wing length, we would expect that individuals with smaller wing lengths would also have smaller body size, and that body size loss would be different. The day of first capture may affect body size decline with age, as individuals emerging later in the flight season may have less time to feed and mate, and may have less and lower quality food available, so body size decline may be faster. For example, females may have less reserves to invest in egg maturation, and males have less time to mate.

The relationship between body mass, thorax width and wing length, and the effect of wing length on body mass and thorax width decline with age were also investigated with these models.

Individual butterfly identifiers nested in year were used as hierarchical random factors (Table 4), as individuals were not independent from certain years. Body mass was log-transformed to improve model fit and to control for initial body mass.

We built these 'full' models with both random slope and random intercept structures. Based on AICc values, the random slope models were more supported for both response variables. Then we removed all non-significant ($p>0.05$) interactions from the full model and then applied an all-combination automated AICc-based model selection from this reduced model using the 'dredge' function of the 'MuMIn' package (Bartoń, 2020). The pre-selected/reduced model for body mass included 11 explanatory variables and 520 models were tested. For thorax width, 13 explanatory variables were included and 1073 models were tested. Finally, parameter estimates of models with ΔAICc<4 were averaged using the 'model.avg' function of the 'MuMIn' package. Model diagnostics of the most supported models (Table 4) were checked by inspecting residual plots; the

‘VarCorr’ function (‘nlme’ package) was used to calculate the proportion of variance explained by the random term.

We also compared the initial mean values for body mass and thorax width between individuals with only one or repeated (at least two) measurements.

4.4.2 Survival analyses

4.4.2.1 Data preparation

We calculated mean daily temperature and relative humidity from data measured between 9 am and 6 pm each day in every ten minutes, and mean nocturnal values between 6 pm and 9 am. Daily and nocturnal variables were strongly correlated (Figure 5), thus we decided to use only daily weather parameters as covariates. As survival probability refers to sampling intervals between consecutive sampling occasions, we used weather variables of the previous day (‘*T_PREV_DAY’; ‘RH_PREV_DAY*’) while for recapture probability, we used weather variables of the given day (‘*T_DAY’; ‘RH_DAY’*) as covariates. When sampling was paused/suspended during the MRR (due to bad weather), we calculated the mean daily temperature and relative humidity of the days without sampling and used them as covariates for survival probability.

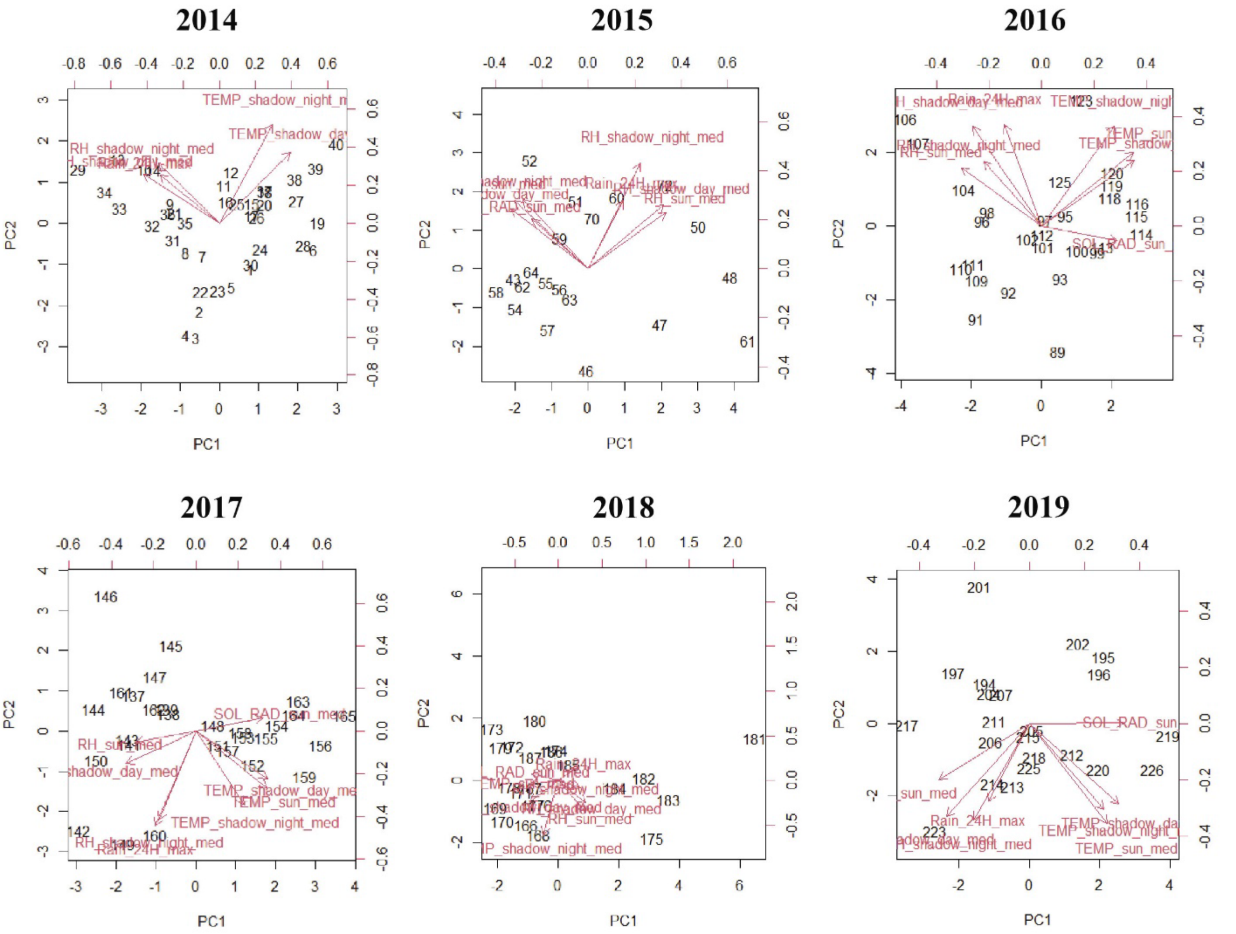


Figure 5: Results of PCA among different weather parameters.

TEMP_shadow_night_med: Median night temperature (between 6 pm and 9 am) measured in the forest surrounding the study area; *RH_shadow_night_med*: Median night relative humidity (between 6 pm and 9 am) measured in the forest surrounding the study area; *TEMP_shadow_day_med*: Median daily temperature (between 9 am and 6 pm) measured in the forest surrounding the study area; *RH_shadow_day_med*: Median daily relative humidity (between 9 am and 6 pm) measured in the forest surrounding the study area; *Rain_24H_max*: Total rainfall in 24 hours; *SOL_RAD_sun_med*: Median daily solar radiation (between 9 am and 6 pm) measured in the open area of the study field

The last 2 days (23rd and 24th of May) of the MRR study in 2014 were omitted from the analysis, because the logger did not measure these days due to technical issues. The dataset of 2020 was omitted from survival analysis, because we had no appropriate weather data for this year.

Temperature and humidity were recorded in sunny spots and also in the shadow in the study site, but these data also correlated, therefore we used only data from loggers in the shadow, because of the more complete dataset. Total rainfall and solar radiation were omitted from analyses because of the incomplete dataset.

‘Day of flight period’ (DOF) was the day in each year when the MRR study was conducted, DOF=1 being the first sampling day of the flight period.

‘Age’ was defined as the number of days elapsed from the first capture of each butterfly. We assumed that all butterflies had been captured soon after their eclosion.

Those individuals that were captured, marked and released at the same sampling day, formed a cohort.

‘DOF’, ‘Age’, and ‘Cohort’ were numeric, continuous variables. With the progress of the flight period, all three variables increased: DOF increased for all individuals, Age increased during the life of individuals, while Cohort denoted different groups of individuals. They were time-varying covariates together with weather variables and the number of observers and they specify whether the given parameter (apparent survival or recapture probability) depends linearly on the day of the flight period or cohort or age (i.e. decreases or increases linearly with the day of the flight period (DOF), with the day of marking (Cohort) or with the days since marking (Age)).

The number of observers was used as a factorial time-varying covariate (‘nobs_lev’) with three levels: (1) only one observer, (2) two or three observers, (3) four or more observers per day were in the field and participated in the MRR study.

We also used ‘time’ as a factor variable that varies among each sampling occasion and it specifies whether apparent survival or recapture probability changed with time. By using ‘time’ as a factorial covariate, we specified such ‘full’ models where apparent survival and/or recapture probability were estimated for each sampling interval/occasion. It is equivalent to that if ‘DOF’ had been a factorial variable.

We used the initial measurements of body mass and thorax width, along with the only measurements of wing and proboscis length (which were assumed to be constant throughout the butterfly's life) as individual covariates in the Cormack-Jolly-Seber (CJS) models. Usually, we measured Apollos on the first occasion when we captured and marked them, except 2014–2016, when the scale was in the field only every third day. Body sizes' quadratic term was also used to model a non-linear relationship between them and the model parameters (apparent survival and recapture probability).

For CJS models, individuals with at least one measurement of all body sizes (body mass, thorax width, wing and proboscis length) were only included, therefore certain butterflies had to be omitted. Almost all years, at least 75% of individuals were included in the CJS models, except for 2014 when only 72.62% of the females' data could be used. In contrast, in 2018 and 2019, more than 98% of the data for both sexes could be included in the analyses (Table 1).

Table 1: The number of individuals in the CJS input files (individuals which had at least one measurement of body mass, thorax width, wing length and proboscis length as well), marked individuals and the percentages of individuals which were included in the CJS analyses.

		no of individuals in the CJS input file	**all marked individuals**	**% of individuals in the CJS analyses**
2014	**males**	103	123	83.74
	females	61	84	72.62
2015	**males**	69	92	75.00
	females	63	84	75.00
2016	**males**	95	106	89.62
	females	78	88	88.64
2017	**males**	81	102	79.41
	females	67	87	77.01
2018	**males**	152	155	98.06
	females	116	116	100.00
2019	**males**	119	120	99.17
	females	82	83	98.80

4.4.2.2 Model selection

We used the Cormack-Jolly-Seber (CJS) model (Cormack, 1964; Jolly, 1965; Seber, 1965) for analysing mark-recapture data. The model has two parameters, apparent survival ('Φ' or '*Phi*') and recapture probability ('*p*'). Apparent survival refers to an interval between two subsequent sampling occasions and it gives the probability that a marked individual which lived at occasion *i* will be alive and available for sampling (i.e. not emigrated) at occasion i+1. Recapture probability refers to sampling occasions, and it gives the probability that marked individuals that are present alive in the population (i.e. available for sampling) at occasion *i* will be captured. Basic assumptions of this model are that (i) all marked individuals have the same recapture probability and (ii) the same survival probability, and (iii) markings are not lost and are read correctly. As we sampled a single, open population, emigration and death could not be distinguished. Nonetheless, the surveyed population was quite isolated, thus we assume that our estimates of apparent survival give a good approximation of real survival.

We aimed to test the relationships between time-varying and individual covariates on apparent survival and recapture probability of the butterflies. Time-varying covariates, i.e. variables that changed during the flight period, were the day of the flight period (DOF), butterfly age (days since marking), cohort (day of marking), the number of observers, and daily mean temperature and relative humidity. In 'Age' models, survival and recapture probability could change linearly over the course of an individual's capture history, which began from the first day of marking. In 'Cohort' models, parameters could change linearly with cohort number. Initial body mass, initial thorax width, wing and proboscis length, as well as their quadratic terms were used as individual covariates, i.e. they had different values among the marked butterflies, but did not change during the flight period.

We used a modified version of the build-up method described in Morin *et al.*, (2020) as a model selection strategy to find those covariates that had a significant relationship with apparent survival and recapture probability. A separate model selection was performed for each year, because generations are not overlapping, and for each sex because of protandry, i.e. early in the flight periods, only males were present in the population, while only females were present at the end. With all these covariates, even without any interactions, we could have built 23653 (109*217) models for each year and each sex. This exceeded our capacity; thus, we limited the number of covariates included in the tested models. In the first step, we included three time-varying covariates, DOF, Age and Cohort, in the models for both parameters and we modelled a linear relationship of the covariates with *Phi* and *p.* To keep the models comprehensive, for each parameter only one covariate was used. In addition, each parameter could be constant, i.e. it had no covariates and it had only one value for the whole flight period. We also defined a fully time-dependent model in which the parameters were estimated for each sampling interval/occasion ('time' models). Thus, each parameter (*Phi* and *p*) could have five covariates and we built 25 models. Most supported models were chosen based on AICc values. We also performed likelihood-ratio (LR) tests on nested models to reveal if DOF, Age and Cohort had significant effects (compared to the constant model).

These first 25 models each year and sex were exported from RMark and imported into the MARK software (White & Burnham, 1999) for Goodness of Fit (GOF) testing. The fit of the models was checked using 3 different methods: i) 'Release GOF' tests the assumptions of CJS model about equal survival and recapture probability among marked individuals, conducting χ^2-tests. Variance inflation factor (VIF or ĉ), which measures overdispersion, was calculated from the ratio of the total χ^2 and the degrees of freedom of deviance. This test is applicable only for the full time-dependent model; ii) for 'Bootstrap GOF-test', simulation of 200 datasets was performed, for Phi(~time)p(~time), Phi(~Age)p(~time) and Phi(~DOF)p(~time) models and models were fit to each simulated dataset. ĉ was calculated in two ways, on the one hand we divided model deviance with mean deviance of simulated datasets, on the other hand we divided model ĉ with mean ĉ of simulated datasets. The highest of the 3 different ĉ values calculated, was considered, in each year, in all three highlighted models and for both sexes.

The full time-dependent models were also tested by U-CARE 3.3 software (Choquet et al., 2009). ĉ was again calculated from the ratio of the total χ^2 and the degrees of freedom of deviance. When ĉ was much greater than one, we ran a ĉ adjustment for the model selection.

Covariates of the most supported models (AICc) and those that had significant effects (LR-tests) were kept for the second step. In the second step, we built models with the covariates carried over from the first step and added the number of observers as a covariate to recapture probability (*p*). We kept the number of observers for the next step if models including it were more supported based on AICc values and/or if LR-tests suggested a significant effect compared to the constant model. In the third step, we again built models with covariates carried over from the previous steps and added mean daily temperature and relative humidity as covariates to both parameters (*Phi* and *p*). As temperature and humidity were clearly negatively correlated (Figure 5), we did not include both for one parameter in one model. Model selection and finding influential covariates were done like in the previous steps. In the fourth step, we built models with covariates carried over from the previous steps and we added body size variables and their quadratic terms. To keep the models simple, we always added one body size variable (and its quadratic term) to the models (Figure 6), and carried out model selection and LR-tests as before. After this, we built models from the most supported covariates from previous steps, but this time we used different body sizes for *Phi* and for *p* (e.g. mass and its quadratic term for *Phi* and wing and its quadratic term for *p*, etc.). We also tested the interactions between wing and proboscis and Age to directly assess whether the effects of body size (wing or proboscis length) and Age on survival are independent, i.e. individuals of different body sizes age similarly. Finally, we summarised the most supported models from all steps into one table and sorted them by their AICc values. We omitted those models that failed to estimate any parameters. We note that the constant models and the fully time-dependent models were included in all steps of the model selection to provide reference models.

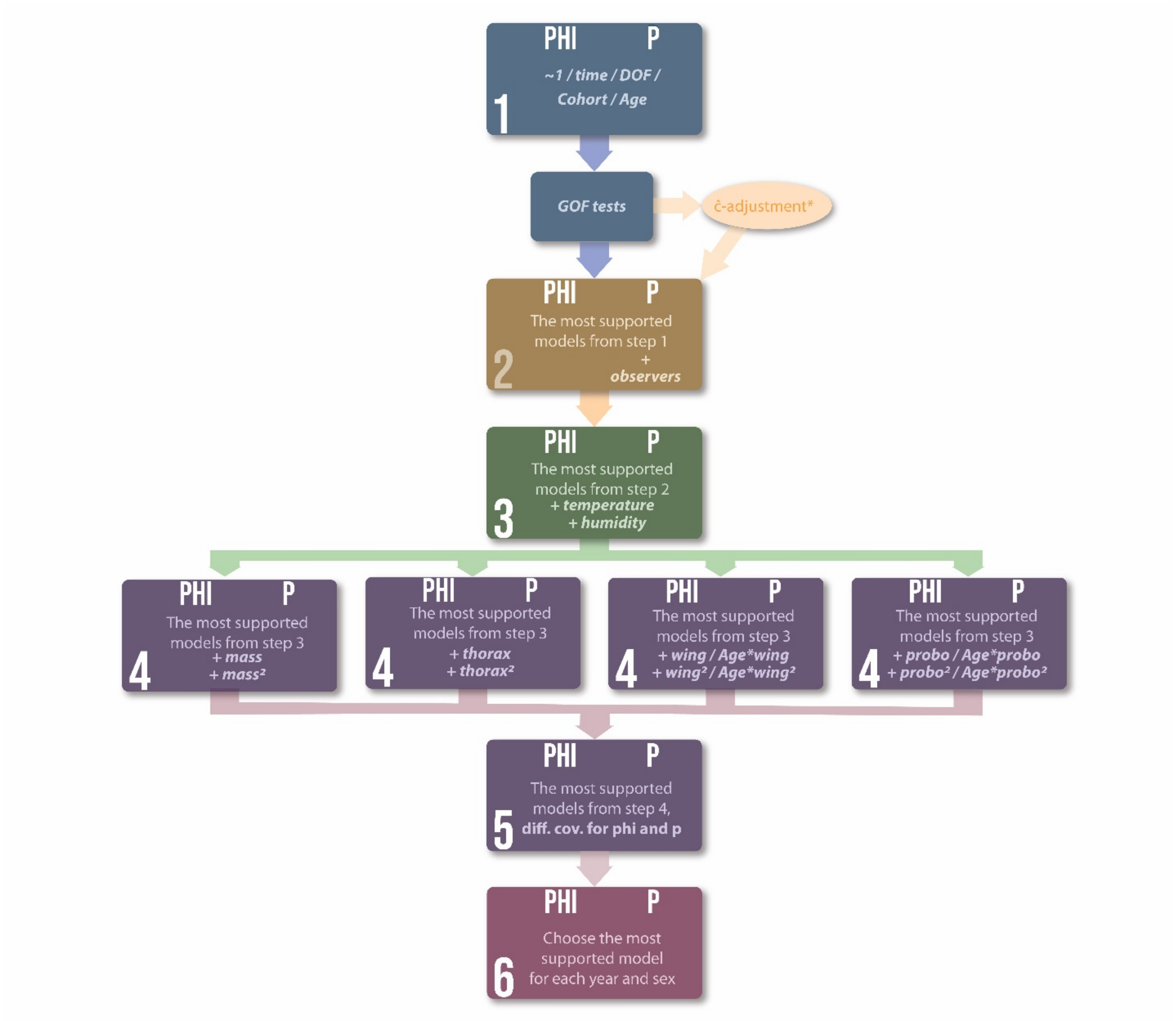


Figure 6: Steps of the model selection of the CJS models. The most supported models were selected based on their ΔAIC values and likelihood ratio tests on nested models. ĉ adjustment was only run for basic models when the results of one of the GOF tests were not appropriate. The models obtained in this way were carried forward to the next steps of the model selection. temperature=mean daily temperature, humidity=mean daily relative humidity, mass=initial body mass, thorax=initial thorax width, probo=proboscis length, wing=wing length.

5. RESULTS AND THEIR DISCUSSION

5.1 Changes and variability in body sizes

5.1.1 Variability in the different body sizes

Between 2014 and 2020, we measured body mass at least once of 1191, and at least twice of 826 (69.35%) Clouded Apollos (Table 2). We measured the thorax width of 1312 individuals, and of 746 (56.86%) individuals repeatedly. The largest proportion of individuals with repeated measures was in 2019 when 82.27% and 80.79% of captured butterflies had repeated body mass and thorax width measurements, respectively. Individuals with the most measurements of body mass (7×, 8× or 9×) occurred in 2016 and 2019, while in the case of thorax width in 2017 and 2019.

Table 2: Number of measured individuals and the number of measurements in each year.

	No. of individuals measured				**No. of individuals with repeated measures (without overlapping)**							
Body mass	**All**	**Single measure**	**Repeated measures**	**% repeatedly measured**	**Number of measurements**							
					2	**3**	**4**	**5**	**6**	**7**	**8**	**9**
2014	167	62	105	62.87	55	32	12	4	2			
2015	138	62	76	55.07	32	23	14	7				
2016	178	32	146	82.02	36	36	27	24	12	10	1	
2017	160	53	107	66.88	52	27	20	8				
2018	271	101	171	63.10	81	59	19	10	2			
2019	203	36	167	82.27	42	48	34	22	9	9	1	2
2020	74	20	54	72.97	22	18	11	2	1			
sum	1191	366	826	69.35	320	243	137	77	26	19	2	2
Thorax width												
2014	205	141	64	31.22	54	8	2					
2015	176	101	75	42.61	60	8	6	1				
2016	193	121	72	37.31	40	21	6	3	2			
2017	189	38	151	79.89	46	49	24	18	11	2	1	
2018	271	106	165	60.89	75	58	19	9	4			
2019	203	39	164	80.79	45	41	33	24	10	9	2	
2020	75	20	55	73.33	19	19	11	5	1			
sum	1312	566	746	56.86	339	204	101	60	28	11	3	

The largest mean initial body mass was in 2019 for females (0.232 g) and in 2020 for males (0.194 g). The smallest was in 2015 for females (0.213 g) and in 2018 for males (0.171 g). The largest mean initial thorax width for both sexes was detected in 2016 (2.131 mm and 2.115 mm for females and males, respectively), and the smallest in 2014 (1.900 mm and 1.851 mm for females and males, respectively) (Table 3).

Table 3: Annual mean ± SE (standard error) initial body mass [g], initial thorax width [mm] and wing length [mm]. Maximum values among years are green, minimum values are orange, the number of individuals is in square brackets. This table contains only those individuals which were included in the Linear Mixed Effects model.

	Mean initial body mass		**Mean initial thorax width**	
Year	**Females**	**Males**	**Females**	**Males**
2014	**0.226** ± 0.005 [64]	**0.179** ±0.003 [103]	**1.90** ± 0.019 [82]	**1.85** ± 0.014 [123]
2015	**0.213** ± 0.006 [67]	**0.173** ± 0.004 [71]	**2.04** ± 0.024 [84]	**1.99** ± 0.017 [92]
2016	**0.227** ± 0.004 [81]	**0.186** ± 0.003 [97]	**2.13** ± 0.018 [88]	**2.12** ± 0.017 [105]
2017	**0.226** ± 0.005 [76]	**0.186** ± 0.004 [84]	**2.09** ± 0.019 [87]	**2.04** ± 0.015 [102]
2018	**0.214** ± 0.003 [116]	**0.171** ± 0.002 [155]	**1.91** ± 0.015 [116]	**1.89** ± 0.014 [155]
2019	**0.232** ± 0.005 [83]	**0.191** ± 0.004 [120]	**2.04** ± 0.016 [83]	**1.97** ± 0.013 [120]
2020	**0.219** ± 0.004 [33]	**0.194** ± 0.004 [41]	**2.03** ± 0.022 [34]	**2.03** ± 0.018 [41]

	Mean wing length	
Year	**Females**	**Males**
2014	**30.69** ± 0.140 [81]	**30.96** ± 0.107 [123]
2015	**30.63** ± 0.154 [84]	**30.75** ± 0.115 [92]
2016	**31.79** ± 0.149 [86]	**31.48** ± 0.110 [103]
2017	**32.31** ± 0.154 [87]	**32.31** ± 0.121 [102]
2018	**31.11** ± 0.113 [116]	**31.11** ± 0.102 [153]
2019	**31.70** ± 0.146 [82]	**31.79** ± 0.120 [119]
2020	**31.36** ± 0.234 [34]	**31.84** ± 0.141 [40]

Body mass and thorax width at first measurements were significantly larger in repeatedly measured individuals (mass: 0.205±0.042 g; thorax: 2.023±0.172 mm; mean±SD) than in individuals measured only once (mass: 0.187±0.038 g; thorax: 1.943±0.193 mm; mean±SD) (mass: $F_{7,1183}$=48.787, p<0.001; thorax: $F_{7,1304}$ =78.852, p<0.001). In repeatedly measured individuals, initial body mass and thorax width were significantly larger for females in all years, but there was no difference observed in wing length between sexes.

The most supported model differed for body mass and thorax width (Table 4); however, random slope models were more supported than random intercept models, indicating individual variance in that the rate of body size declined with age.

Table 4: The full and the most supported models' specifications.

	Response variable	**Explanatory variables**	**Random term**
Linear Mixed-Effects Models (full model)	**Log(Body mass)**	sex*(age + mean age + age^2 + mean_age^2 + first capture + mean first capture + wing) + (age:wing) + (age:first capture)	random=~1|year/id *or* random=~age|year/id
	Thorax width	sex*(age + mean age + age^2 + mean_age^2 + first capture + mean first capture + wing) + (age:wing) + (age:first capture)	random=~1|year/id *or* random=~age|year/id
Most supported models	**Log(Body mass)**	sex*age^2 + wing + age*first capture	random=~age|year/id
	Thorax width	sex*age + mean age + age^2 + first capture + wing	random=~age|year/id

5.1.2 Changes in body sizes with age - Phenotypic senescence

5.1.2.1 Body mass change with age

We found a significant decline in body mass with increasing age in both sexes. This relationship was non-linear, body mass decreased slower at an older age (Figure 7).

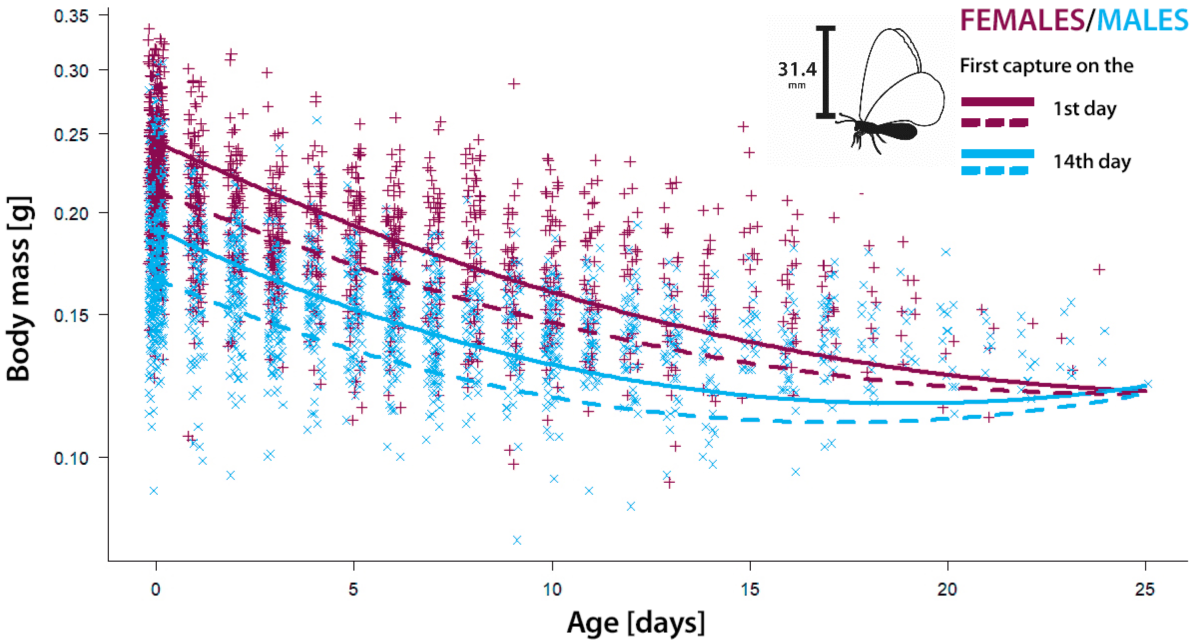


Figure 7: Body mass changed with age. Symbols represent the individuals' measurements (purple+ =females; blue× =males). Dots are slightly jittered along the x-axis for better visibility. The lines illustrate the estimated relationship between age and body mass for each sex, and for an individual with an average wing length of 31.4 mm. Solid lines denote individuals captured first on the 1st day of the flight period, while dashed lines represent those captured first on the 14th day.

Variation and change in the butterflies' body mass can be influenced by many factors. Body mass loss was related to water loss during desiccation in Heliconiinae butterflies; total body water differed among species and it was lower for females than males likely due to higher fat content (Mazer & Appel, 2001). In freshly emerged Woodland Brown butterflies (*Pararge aegeria*), (i) lipid reserves were higher than in the 2–3 days old indicating a decline in lipid reserves with ageing in a laboratory study, and (ii) males with lower lipid reserves formed smaller spermatophores showing a less steep decline in body mass (Vande Velde et al., 2013). Oviposition or spermatophore transmission may also result in the change of body mass (Stjernholm & Karlsson, 2000). Clouded Apollo females may receive sphragis during mating that blocks the copulatory opening and makes up 3–5% of the measured female body mass (Gór, Fónagy, et al., 2023). This presumably causes body mass loss in males and it may also be a burden for females as flying with sphragis could be costly. In addition, post-emergence body mass may be higher due to yet unreleased meconium.

Body mass also significantly declined with the date of first capture, i.e. individuals captured and marked later in the flight period were smaller, but their body mass also declined slower (Figure 7). Finally, body mass was positively related to wing length, but the decline of body mass with age was not affected by wing length (Table 5Table 6).

Table 5: The results of the model selection for log(body mass) for the Linear Mixed-Effects Model with random slope.

Model specification	**AICc**	**ΔAICc**	**df**	**logLik**
log(body mass) ~ sex * age^2 + age * first capture + wing, random=~age|year/id	-4279.6	0.00	15	2154.858
log(body mass) ~ sex * age^2 + mean age + age * first capture + wing, random=~age|year/id	-4279.4	0.17	16	2155.781
log(body mass) ~ sex * age^2 + age * first capture + wing + mean(first capture), random=~age|year/id	-4278.8	0.71	16	2155.512
log(body mass) ~ sex * age^2 + mean age + age * first capture + wing + mean(first capture), random=~age|year/id	-4277.0	2.55	17	2155.604
log(body mass) ~ sex * age^2 + age * first capture + mean age^2 + wing, random=~age|year/id	-4270.4	9.15	16	2151.291
log(body mass) ~ sex * age^2 + age * first capture + mean age^2 + wing + mean first capture, random=~age|year/id	-4268.3	11.24	17	2151.262

After model averaging, the coefficients of mean(age), mean(age^2) and mean(first capture) were not significant. In this kind of model parametrisation, these coefficients are the differences between the within-subject and between-subject effects (Van de Pol & Wright, 2009), thus we can conclude that these effects did not differ significantly. This means that body mass declined significantly with age and that individuals measured at (on average) an older age had lower body mass. We note that the coefficient of mean(first capture) had a relatively large positive value and it was significant in those models where it was included (Table 6), suggesting that the between-subject effect was positive. This means that within each year, individuals captured later had lower body mass, but the average body mass was higher in those years when the flight period was longer (i.e. mean(first capture) was higher). This, however, does not seem to be a very strong relationship.

Table 6: The results of model averaging for log(body mass) for the Linear Mixed-Effects Model (subset=ΔAICc < 4).

Trait	**Model-averaged coefficients (full average)**	**Estimate**	**SE**	**Adjusted SE**	***z*-value**	***p*-value**	
Log(Body mass)	(Intercept)	-2.9810	0.1272	0.1273	23.418	< 0.001	***
	age	-0.0519	0.0019	0.0019	26.843	< 0.001	***
	age^2	0.0009	0.0001	0.0001	13.042	< 0.001	***
	first capture	-0.0103	0.0008	0.0008	13.031	< 0.001	***
	sex(male)	-0.2482	0.0078	0.0078	31.694	< 0.001	***
	wing	0.0497	0.0027	0.0027	18.386	< 0.001	***
	age:first capture	0.0005	0.0001	0.0001	6.530	< 0.001	***
	age^2:sex(male)	0.0004	0.0001	0.0001	7.282	< 0.001	***
	mean(age)	0.0019	0.0023	0.0023	0.789	0.430	
	mean(first capture)	0.0057	0.0081	0.0083	0.687	0.492	

5.1.2.2 Thorax width change with age

Thorax width also declined with age non-linearly in both sexes, but the rate of decline was slightly higher in females. Thorax width was smaller in males than in females and it was positively related to wing length (Figure 8).

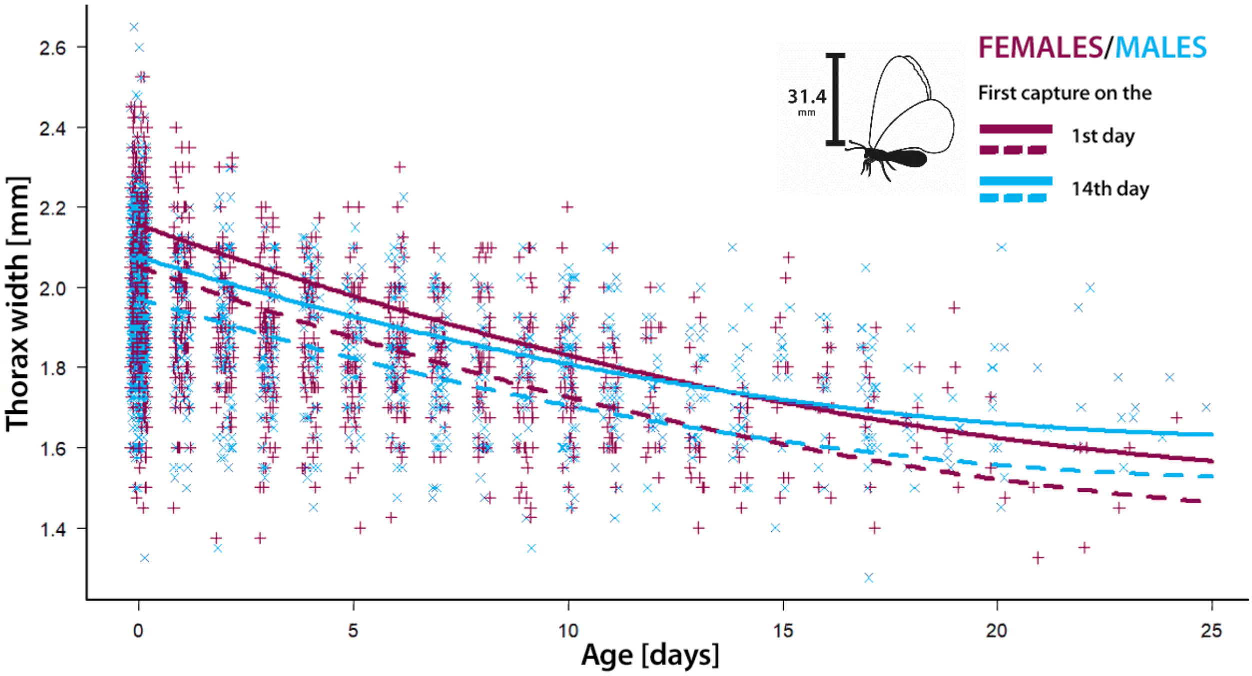


Figure 8: Thorax width changed with age. Symbols represent the individuals' measurements (purple+ =females; blue× =males). Dots are slightly jittered along the x-axis for better visibility. The lines illustrate the estimated relationship between age and thorax width for each sex, and for an individual with an average wing length of 31.4 mm. Solid lines denote individuals captured first on the 1st day of the flight period, while dashed lines represent those captured first on the 14th day.

Butterflies that were first captured later during the season had smaller thorax width. Mean(age) proved to be significant, but its value was relatively low implying that within-subject and between-subject effects had similar directions (Figure 8, Table 7Table 8). This means that thorax width significantly declined with age (within-subject effect) and individuals measured at (on average) an older age had smaller thorax width (between-subject effect).

Table 7: The results of the model selection for thorax width for the Linear Mixed-Effects Model with random slope.

Model specification	**AICc**	**ΔAICc**	**df**	**logLik**
thorax width ~ sex * age + mean age + wing, random=~age|year/id	-3391.3	0.00	15	1710.758
thorax width ~ sex * age + mean age + wing + mean(first capture), random=~age|year/id	-3389.2	2.14	16	1710.697
thorax width ~ sex * age^2 + mean age + age * first capture + wing + age:first capture + age:sex, random=~age|year/id	-3382.9	8.42	16	1707.559
thorax width ~ sex * age^2 + mean age + age * first capture + wing + mean(first capture) + age:first capture + age:sex, random=~age|year/id	-3381.0	10.34	17	1707.613
thorax width ~ sex * age^2 + mean age + age * first capture + wing + mean(first capture) + age:sex + mean(first capture):sex, random=~age|year/id	-3380.6	10.70	17	1707.429

Table 8: The results of model averaging for thorax width for the Linear Mixed-Effects Model (subset=ΔAICc < 4).

Trait	**Model-averaged coefficients (full average)**	**Estimate**	**SE**	**Adjusted SE**	***z*-value**	***p*-value**	
Thorax width	(Intercept)	0.7892	0.1621	0.1621	4.868	< 0.001	***
	age	-0.0386	0.0020	0.0020	19.723	< 0.001	***
	age^2	0.0006	0.0001	0.0001	7.441	< 0.001	***
	first capture	-0.0074	0.0005	0.0005	14.063	< 0.001	***
	mean(age)	0.0080	0.0013	0.0013	6.173	< 0.001	***
	sex(male)	-0.0751	0.0079	0.0079	9.501	< 0.001	***
	wing	0.0397	0.0026	0.0026	15.384	< 0.001	***
	age:sex(male)	0.0059	0.0010	0.0010	6.141	< 0.001	***
	mean(first capture)	0.0059	0.0106	0.0109	0.545	0.585	

We detected high individual variation in initial body sizes and minor variation in its change with age. Random slope models were more supported than random intercept models, indicating that the rate of body size decline with age has a non-negligible individual variance. The random factors explained a high proportion of the total variance in the most supported models for body mass (~70%) and thorax width (~62%) (Table 9).

Table 9: Proportion of variance of random terms in Linear Mixed-Effect Models

Trait	Random effects	Fixed effects	Variance	Proportion of variance (%)
Body mass	id	Intercept	0.0118	50.40
		age	0.000035	0.15
	year	Intercept	0.0045	19.22
		age	0.00001	0.04
	Residual		0.00707	30.19
Thorax width	id	Intercept	0.0088	31.17
		age	0.000019	0.07
	year	Intercept	0.0086	30.46
		age	0.000014	0.05
	Residual		0.0108	38.25

The highest proportion of variance was explained by the between-individual differences in both variables. In case of body mass, this variation was much higher than between-year variance, while for thorax width the between-year variation was nearly the same as the between-individual variation. These indicate that initial body mass had a larger between-individual than between-year variation, while initial thorax width had similar variation between individuals and between years. The age-effect showed much lower variation indicating that initial body mass and thorax width had much higher variation than the rate of ageing.

Clouded Apollos spend a lot of time feeding (Szigeti et al., 2018b). In our study population, individuals' body mass may vary ~10% daily with the timing of feeding vs. other activities (flying; patrolling; mating; egg-laying) that incur net weight loss. These activities may cause detectable changes in body mass within a day and might explain the high individual variation.

Reserves used from abdomen and/or thoracic muscles may cause a decline, especially in thorax width. Resource allocation during adult and (possibly) larval stages can be affected by trade-offs between survival and reproduction (Boggs, 2009). For instance, nitrogen content of the flight muscles may be used for reproduction in the Green-veined White (*Pieris napi*) butterfly females (Karlsson, 1998), causing thorax mass decrease. However, using thoracic muscles as a resource may be restricted by flight performance (Stjernholm et al., 2005). This may be the case of Clouded Apollo males as they should continuously patrol to find new mates, in the first half of the flight period because of male bias in the adult sex ratio, in the second half of the flight period because of the significant proportion of females that have sphragis and virgin females are harder to find (Gór, Lang, et al., 2023).

5.1.3 Sexual Dimorphism in body size changes

We observed sexual dimorphism in body size changes, characterized by a higher rate of decline in females in terms of the rate of change. While males initially had significantly lower body mass than females, the decline in males' body mass was slower at an older age, as indicated by the significant sex*age^2 interaction. Consequently, both sexes exhibited approximately similar body mass by the end of their lifespan.

Both initial body mass and thorax width were larger in females. Body mass declined slower with age in males only at an older age (Figure 7). Conversely, thorax width showed a slower decline in males even at a younger age, resulting in males having, on average, wider thoraces than females after approximately 14 days of age (Figure 8). The presence of such differences may be influenced by many circumstances and can be derived from the dissimilar physiology and behaviour of the sexes. Accelerated decline in body mass or thorax width may be caused by egg-laying in females and by producing spermatophores and/or sphragides in males. Resource allocation can be different between the sexes, e.g. females may use more nutrients from muscle breakdown from the thorax for reproduction than males, therefore experiencing a steeper decline (Stjernholm et al., 2005), but males may also invest into reproduction through the spermatophore, and additionally in the case of the Clouded Apollo, through the sphragis (Gór, Lang, et al., 2023; Vlašánek & Konvička, 2009). Flying is energetically costly (Dudley, 2002) and male butterflies often fly more than females (e.g. Popović et al., 2022). According to our field observations, Clouded Apollo males spend a lot of time patrolling to find mating partners, hence they possibly lose more water and reserves from their body than females. Moreover, as male reproductive success likely depends on flight ability more than females', there might be a strong selection against decomposing their thoracic muscles. In contrast, 69–77% of females are sphragis-bearing (Gór, Fónagy, et al., 2023), and flight costs might be higher for them than for those lacking sphragis or for males. We have no data on the frequency of feeding and the amount of nectar consumed, but it is possible that either males or females can compensate for the faster body mass loss with a higher feeding rate.

5.1.4 Variability in initial body sizes

Body size of individuals newly appearing in the population showed a declining trend with the progress of the flight period. Clouded Apollos are protandrous (Gór, Lang, et al., 2023), i.e. males emerge earlier in the flight period than females to maximize their reproductive success (Fagerström & Wiklund, 1982; Wiklund & Fagerström, 1977). A recent meta-analysis in insects revealed that the direction and degree of sexual bimaturism and sexual size dimorphism are positively associated (Teder et al., 2021). This is partly supported by our results as females were larger than males in all years. Unlike in a laboratory study, here we could not control for environmental conditions (e.g. temperature, humidity, food availability etc.) during larval development, therefore we can only speculate about the reasons underlying our results. Based on the fact that larger body size usually means higher fecundity in insects (Honěk, 1993) and that later emergence has no apparent benefit, we suspect that there is a 'growth race' among the larvae and individuals pupate upon reaching an optimum size. This optimum might be influenced by genetic factors as well as environmental constraints on development. Winners of the growth race, i.e. the early pupating and emerging individuals, must be soon followed by slower developers, otherwise they would not find mating partners. Thus, the slower developing individuals might be forced to pupate before reaching the body size of earlier individuals. Note that classical life history theory typically predicts a decreasing reaction norm between optimal body size and age at maturity when growth rate varies in different environmental conditions (Stearns & Koella, 1986).

Alternatively, Clouded Apollos appeared later in the flight period, may have hatched later from the eggs, so they may have had less time to develop. Later hatching can also be disadvantageous because aged host plants may have lower nutrient content (e.g. Mattson, 1980) and/or higher concentration of defence chemicals or physical defences (Barton & Koricheva, 2015; Yang et al., 2020). Restricted amount or bad quality food may also cause prolonged larval development time (Gibbs et al., 2012) or smaller adult body size (Boggs & Freeman, 2005; Boggs & Niitepõld, 2016; Niitepõld & Boggs, 2022).

Finally, the mean age of butterflies increases with the progress of the flight period. This means that unmarked individuals captured later in the season may indeed have had a higher age than individuals captured first earlier in the season (the exact age of unmarked individuals could not be assessed).

We revealed that butterflies with repeated measurements had significantly higher initial body mass and thorax width than those measured only once, i.e. we encountered, captured and measured larger Clouded Apollos more often than the smaller ones. This is likely due to the fact that butterflies appearing earlier in the flight period were larger and had a higher chance to be recaptured and measured multiple times. We also found that butterflies with longer wings had higher initial body mass, but wing length did not affect body mass decline. Individuals with wider thoraces and longer wings may indicate better flight ability and greater activity levels (Altizer & Davis, 2009; Davis & Holden, 2015; Sekar, 2012; Soule et al., 2020), leading to higher detectability and/or better survival abilities, but further investigations are needed to clarify this phenomenon.

5.2 Survival analyses – Actuarial senescence

5.2.1 Variation in the measured variables

The longest flight period was 45 days in 2019, while the shortest with 26 days was in 2018, when the most Clouded Apollos (271) were marked. 2018 was the only year when no days were omitted from sampling, all other years had missed days due to bad weather conditions (Table 10).

Table 10: Dates, duration (days), the number of sampling occasions (days) and the number of marked individuals in each flight period.

Year	First day	Last day	Duration	Sampling occasions	N_{males}	$N_{females}$	$N_{individuals}$
2014	2014-04-17	2014-05-26	40	33	123	84	207
2015	2015-04-26	2015-05-30	35	29	92	84	176
2016	2016-04-22	2016-06-03	43	34	106	88	194
2017	2017-04-25	2017-05-29	35	34	102	87	189
2018	2018-04-29	2018-05-24	26	26	155	116	271
2019	2019-04-21	2019-06-04	45	34	120	83	203

During the flight periods in every year, both temperature and relative humidity fluctuated (Table 11, Appendix 2Appendix 6). The lowest daily mean temperature was measured in 2016 (5 °C), while the highest was in 2015 (26.7 °C). The lowest and the highest relative humidity (24.59% and 99.30 %) were both in 2019.

Table 11: Minimum, mean and maximum daily (between 9 am and 6 pm) temperature and relative humidity for each year.

	Temperature [°C]			Relative humidity [%]		
	Min.	**Mean**	**Max.**	**Min.**	**Mean**	**Max.**
2014	9.80	16.41	25.35	45.25	74.31	97.65
2015	12.10	18.77	**26.70**	40.95	64.10	97.60
2016	**5.00**	18.34	24.90	40.70	75.27	94.00
2017	7.40	17.60	23.60	47.00	74.10	93.00
2018	13.55	22.84	24.95	40.10	69.95	95.05
2019	5.50	23.42	25.90	**24.59**	75.27	**99.30**

The largest mean initial body masses occurred in 2019 (0.232 g and 0.191 g for females and males, respectively), the widest mean initial thoraces occurred in 2016 (2.15 mm and 2.11 mm for females and males, respectively), while the longest wings (32.43 mm and 32.30 mm for females and males, respectively) and proboscides (12.24 mm and 12.18 mm for females and males, respectively) were measured in 2017 for both sexes (Table 12).

The lowest female initial body mass was in 2015 (0.212 g) and for males in 2018 (0.172 g), while the smallest female initial thorax width was measured in 2018 (1.91 mm) and for males in 2014 (1.87 mm). The shortest wing length was in 2015 for both sexes (30.6 mm and 30.72 mm for females and males, respectively), while the shortest proboscis length was in 2018 for both sexes (11.62 mm and 11.67 mm for females and males, respectively) (Table 12).

Table 12: Annual mean ± SE initial body mass [g], initial thorax width [mm], wing length [mm] and proboscis length [mm]. Maximum values for each year are green, minimums are orange, the number of individuals is in square brackets. This table contains only those individuals which were included in the CJS analyses.

	Mean initial body mass		**Mean initial thorax width**	
Year	**Females**	**Males**	**Females**	**Males**
2014	**0.226** ± 0.005 [61]	**0.179** ± 0.003 [103]	**1.93** ± 0.022 [61]	**1.87** ± 0.016 [103]
2015	**0.212** ± 0.006 [63]	**0.173** ± 0.004 [69]	**2.06** ± 0.027 [63]	**2.00** ± 0.019 [69]
2016	**0.227** ± 0.005 [78]	**0.186** ± 0.003 [95]	**2.15** ± 0.018 [78]	**2.11** ± 0.017 [95]
2017	**0.229** ± 0.005 [67]	**0.187** ± 0.004 [81]	**2.12** ± 0.019 [67]	**2.06** ± 0.015 [81]
2018	**0.214** ± 0.003 [116]	**0.172** ± 0.002 [152]	**1.91** ± 0.015 [116]	**1.88** ± 0.013 [152]
2019	**0.232** ± 0.005 [82]	**0.191** ± 0.003 [119]	**2.05** ± 0.016 [82]	**1.97** ± 0.013 [119]

	Mean wing length		**Mean proboscis length**	
Year	**Females**	**Males**	**Females**	**Males**
2014	**30.79** ± 0.162 [61]	**31.00** ± 0.110 [103]	NA	NA
2015	**30.60** ± 0.184 [63]	**30.72** ± 0.128 [69]	**12.20** ± 0.075 [63]	**12.11** ± 0.074 [69]
2016	**31.89** ± 0.150 [78]	**31.48** ± 0.118 [95]	**12.07** ± 0.071 [78]	**12.08** ± 0.052 [95]
2017	**32.43** ± 0.173 [67]	**32.30** ± 0.144 [81]	**12.24** ± 0.071 [67]	**12.18** ± 0.072 [81]
2018	**31.11** ± 0.113 [116]	**31.12** ± 0.102 [152]	**11.62** ± 0.046 [116]	**11.67** ± 0.043 [152]
2019	**31.70** ± 0.146 [82]	**31.79** ± 0.120 [119]	**11.87** ± 0.078 [82]	**11.95** ± 0.052 [119]

The most supported models (Table 13, Appendix 3–4) were not the same for all years, but the directions of the covariates’ effects were similar.

Based on the results of GOF tests (Appendix 5) we performed ĉ-adjustment on the basic models in 2017 and 2019 for males, and in 2018 for both sexes, because the estimation of Bootstrap GOF and U-CARE were too high in these cases (Appendix 5). Despite the adjustment the significance of the variables (Age, DOF, Cohort, time) and the order of the most supported models were not changed. Regarding the Release GOF test, GOF statistics (TEST 2 and TEST 3) were significant for some sampling occasions in 2017 and 2019 for males and in 2018 for both sexes (Appendix 6). In Release GOF, TEST 2 focuses on animals confirmed to be alive between occasions (i) and (i+1). It requires individuals marked on or before occasion (i) and those captured on or after occasion (i+1). If an individual was marked at (i) and recaptured at (i+1) or later, it indicates the animal was alive during this interval. The objective of this analysis is to determine whether the recapture probability is dependent on the specific occasion on which the butterfly was marked. TEST 3 examines whether all marked animals alive at occasion (i) have an equal probability of surviving to occasion (i+1). It investigates that from the individuals seen at occasion (i), how many were seen again, and which occasion. This test considers whether being seen for the first time at occasion (i) or prior to it affects the recapture probability of (TEST.SR) or the time of the recapture (TEST3.Sm)

The significant findings indicate that there was some heterogeneity in both survival probability and catchability among the individuals on these sampling occasions. Temporary emigration of males could be an explanation, or in case of females, difficulties in recapture might be attributed to mating or egg-laying activities in the grass. On certain sampling occasions, when a large number of butterflies were captured simultaneously, they were temporarily held in envelopes. Delayed release, such as after several hours or the following morning, may impact recapture probability, as the butterflies may require additional time to warm up and become fully active for flight.

The observed heterogeneity in survival rates could be explained by variations in circumstances among the individuals during these years (e.g. difference in weather conditions, nectar plants availability or body sizes). However, we emphasise that less than 5% of the sampling occasions showed such discrepancies in the Release-GOF tests.

Apparent survival declined with age or with the day of the flight period in every year and for both sexes. In most years, survival was negatively related to temperature and positively to relative humidity. And finally, in some years body size was significantly positively related to survival (which means that larger individuals had a higher survival probability).

In most of the years, the most supported models for recapture probability were 'time' models, so recapture probability varied among sampling occasions within these years and the covariates could not explain its variance sufficiently. In some years, recapture probability was significantly higher when more observers worked in the field, when temperature was higher and relative humidity was lower. In some other years, recapture probability increased with age and body sizes, i.e. older and/or larger individuals were recaptured with higher probabilities.

Not only those covariates that were included in the most supported models were significant (for all significant covariates see). Likelihood ratio tests resulted in several variables being significant in certain years but not included in the most supported model. This is weaker evidence for an effect than for the variables included in the most supported model.

Table 13: Overview of the most supported models of the Cormack-Jolly-Seber model selection. These models were selected based on their AICc values and likelihood ratio tests. Blue or red indicates negative or positive effects of variables on apparent survival (Phi) or on recapture probability (p). DOF: Day of the flight period, mass: initial body mass, thorax: initial thorax width, wing: wing length.

Year	**Sex**	**Apparent survival (Phi)**	**Recapture probability (p)**
2014	**males**	~Age + temperature	~time
	females	~DOF + relative humidity	~relative humidity + thorax + thorax^2
2015	**males**	~DOF + thorax	~observers + Cohort + wing + wing^2
	females	~DOF	~observers + temperature + thorax
2016	**males**	~DOF + temperature	~time
	females	~Age + temperature	~Age + relative humidity
2017	**males**	~DOF + relative humidity	~time
	females	~DOF + relative humidity	~time
2018	**males**	~DOF	~Cohort + relative humidity
	females	~DOF	~time
2019	**males**	~Age + temperature + wing	~time
	females	~Age + temperature + mass	~temperature

Circumstances (like weather and available resources, e.g. nectar plants) experienced by individuals were changing from year to year in this natural population (Szigeti et al., 2018a), therefore the effects of variables were also different. In the following sections the direction and degree of the variables' effects from the most supported models are presented. Note that figures were made either from the most supported models or from other models where the given covariate was significant, and the y-axes are set to different scales, in order to better represent the direction of the relationships. Several covariates were significant in the most supported models in general, so it is not easy to plot the effect of a single covariate in the figures. In some of the following figures, only little differences can be seen in the survival probabilities. To interpret these differences, it is worth calculating the average lifespan (e). In principle, this can be estimated using the following formula:

$$e = (1 - \Phi)^{-1} - 0.5$$

With this formula average life span (e) can be calculated from the daily survival rate (Φ) (Bubová et al., 2016; Nowicki et al., 2005). Note, that this formula can only be used for constant survival probabilities.

For instance, when the daily survival rate is 0.9, it implies an average lifespan of 9.5 days, while when the daily survival rate is 0.8, it implies a lifespan of only 4.5 days. This example only illustrates that what may appear to be a small difference in the figures of survival, in reality gives a large difference in average lifespan. However, this formula is only applicable if the survival probability is constant, which in our case is not true, so no further lifespan has been calculated, the above example is for illustrative purposes only.

5.2.2 Relationship between Age and apparent survival (actuarial senescence) and between Age and recapture probability

Age had a significant negative effect on apparent survival in all years and for both sexes meaning that survival probability declined with increasing age, except in 2018, when Age was not significant. Age was included in the most supported models in 2019 for both sexes, in 2016 for females (Figure 9) and in 2014 for males (Figure 10).

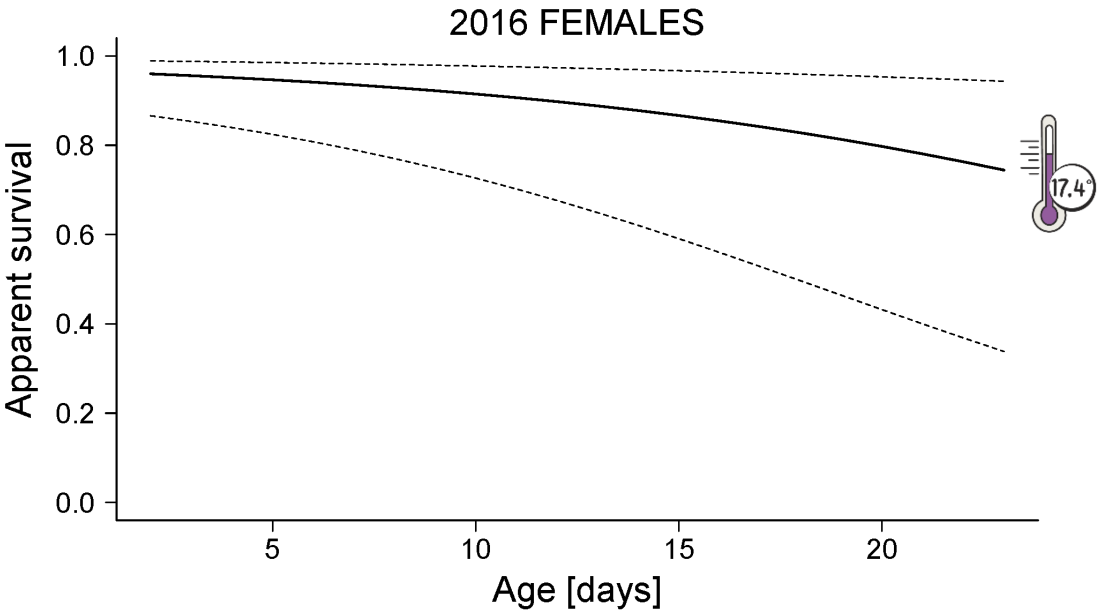


Figure 9: Estimated mean apparent survival for females (Phi(~Age+temperature)p(~Age+relative humidity) model, median temperature in 2016 = 17.4 °C) in relation to the number of days since marking (Age) in 2016. Dashed lines represent 95% confidence intervals.

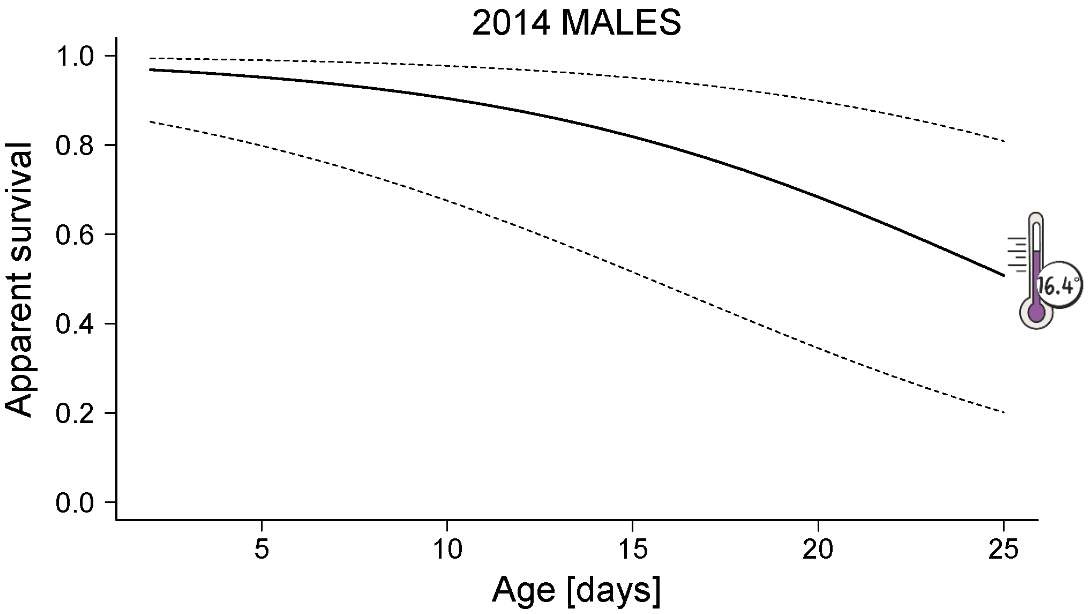


Figure 10: Estimated mean apparent survival for males (Phi(~Age + temperature)p(~time) model, median temperature in 2014 = 16.4 °C) in relation to the number of days since marking (Age) in 2014. Dashed lines represent 95% confidence intervals.

Our study is one of the few (Carroll & Sherratt, 2017; Sielezniew et al., 2020) to detect actuarial senescence in a natural butterfly population. According to our results survival significantly declined with age in five out of six consecutive years for both sexes. Our findings are consistent with the results of Sielezniew et al. (2020), that initial apparent survival was slightly different between males and females, in some years, the males aged faster, while in other years, the females did (Appendix 4).

Longer-lived individuals often have extended windows for reproductive activities, allowing them to potentially invest more in reproduction throughout their lives (Boggs & Freeman, 2005; Fischer, 2007; Kőrösi et al., 2008), and they have more time to feed and store resources. In case of Clouded Apollos, longer-lived individuals may have more time to mate, males may have more opportunities to mate with several females and females may lay more eggs. Female Clouded Apollos lay eggs multiple times throughout their lives, individually ovipositing them across various surfaces and locations, making it a time-consuming act (Gór, Fónagy, et al., 2023; Gór, Lang, et al., 2023; Wiklund, 1984). Therefore, a longer lifespan may imply a higher fecundity (see also Kőrösi *et al.*, 2008).

Age had a significant positive effect on recapture probability only in 2016 for females (Figure 11), and it was also included in the most supported model in this year. It means that recapture probability was higher in older female butterflies.

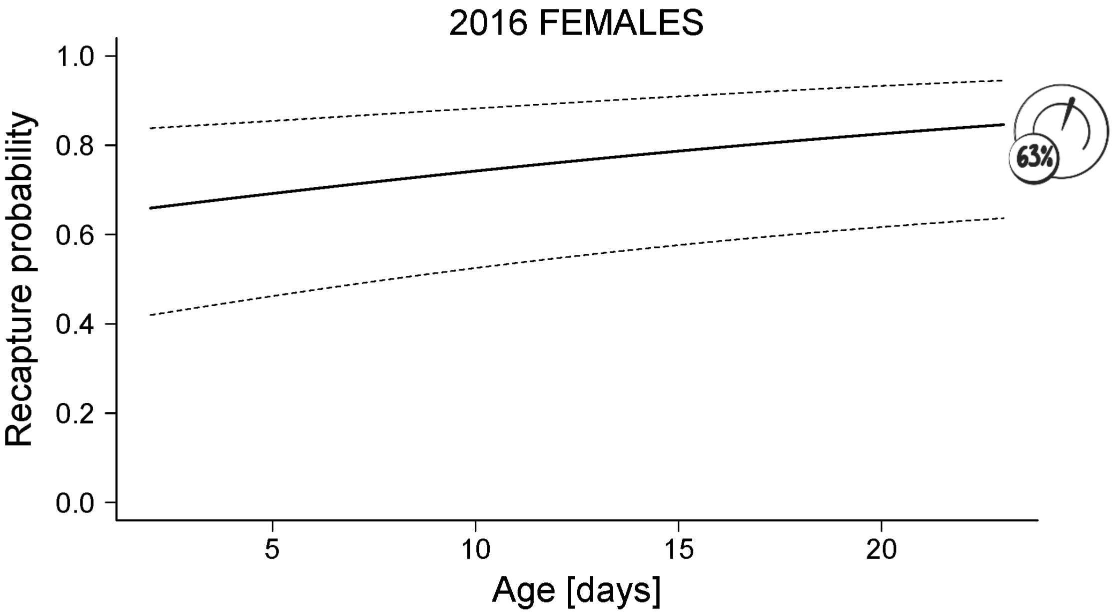


Figure 11: Estimated mean recapture probability for females (Phi(~Age+temperature)p(~Age+relative humidity) model, median relative humidity in 2016 = 63%) in relation to the number of days since marking (Age) in 2016. Dashed lines represent 95% confidence intervals.

Several factors affect the detectability of individuals. Presumably, older butterflies are easier to observe due to reduced activity levels, for example, when they are basking with their wings open on vegetation (dorsal basking; Kingsolver, 1985; Wickman, 2009). Physiological deterioration, such as thorax muscle atrophy as suggested by Stjernholm, Karlsson and Boggs, 2005 (see also Section 5.1) and declining energy reserves likely contribute to this decreased activity, resulting in reduced energetically demanding behaviours such as flight or foraging in older individuals. Alternatively, older females may fly weaker and thus more likely to be captured.

Conversely, observers might also detect more active individuals that fly frequently and are more visible with a higher probability.

5.2.3 Relationship between the Day of the flight period and apparent survival, the Day of the flight period and recapture probability, and between Cohort and apparent survival, and Cohort and recapture probability

The day of the flight period was significantly negatively related to apparent survival (Figure 12) in all years and both sexes, meaning that survival probability declined with the progress of the flight period. It was included in the most supported models in 2015, 2017, 2018 for both sexes, in 2016 for males and in 2014 for females.

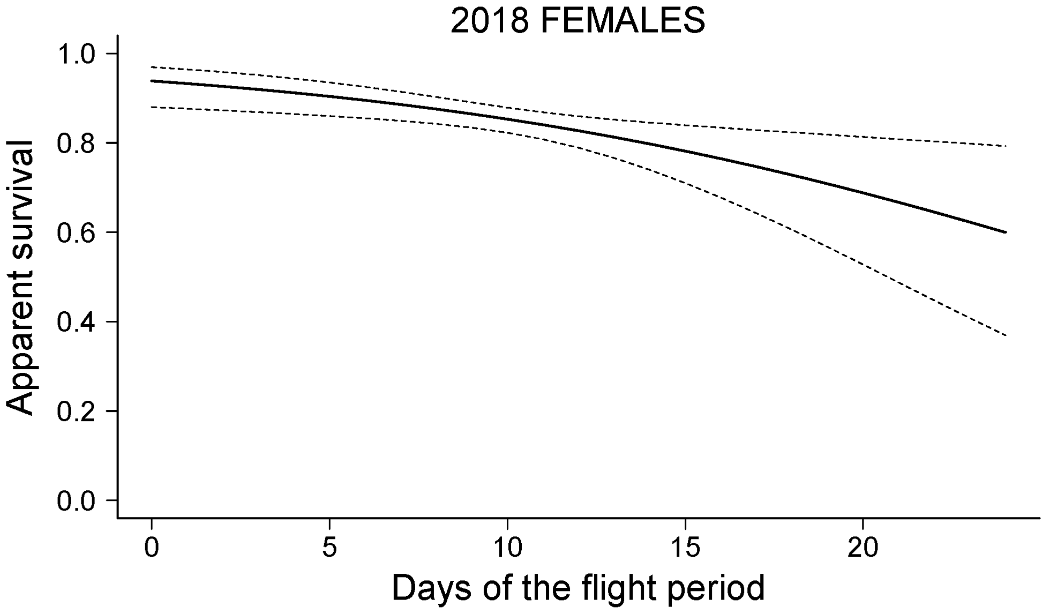


Figure 12: Estimated mean apparent survival for females (Phi(~DOF)p(~time)model) in relation to the day of the flight period (DOF) in 2018. Dashed lines represent 95% confidence intervals.

The day of the flight period was significantly negatively related to recapture probability in 2014, 2016, 2017 and 2018 for males, and had a significant positive effect in 2019 for females, however it was not included in the most supported models in any of the years.

A higher proportion of older individuals in the second half of the season, known for their lower survival probability, might have contributed to the overall decline in apparent survival. Moreover, individuals which emerge later in the flight period may experience suboptimal conditions with reduced resource availability e.g. nectar plants which may negatively affect survival (Cahenzli & Erhardt, 2012), or increased competition for resources. Besides that, increasing temperature with the progress of the flight period may lead to an increased metabolic rate, accelerating ageing and resulting in a decline in survival probability (Niitepõld, 2010). Although our study population was rather closed in terms of movement, we cannot exclude that the emigration propensity of butterflies increased with the progress of the flight period.

An opposite relationship was observed for males and females between the day of the flight period and recapture probability, although for females this relationship was significant in only one year. Vlašánek and colleagues observed a behavioural switch in several butterfly species. As the flight period progresses, males' transition from maintenance to reproduction (patrolling), while females shift from reproduction to maintenance. Consequently, oviposition and resting in females decreased, typically towards the end of the season (Vlašánek et al., 2018). In contrary, our results showed that males' recapture probability decreased with the progress of the flight period, probably because of ageing or because smaller freshly emerged males patrol less and spend more time hiding in the vegetation. However, the recapture probability of females increased with the progress of the flight period, possibly because they spent more time feeding, making them more conspicuous.

Cohort was significantly negatively related to apparent survival, but only for males in 2016 and 2018, and it was not included in the most supported models in any of the years.

Cohort was significantly negatively related to recapture probability for males in 2015, 2017 and 2018, and had a significant positive effect for females in 2019 (in the same year when DOF had a positive effect). It was included in the most supported models for males in 2015 and 2018 (Figure 13).

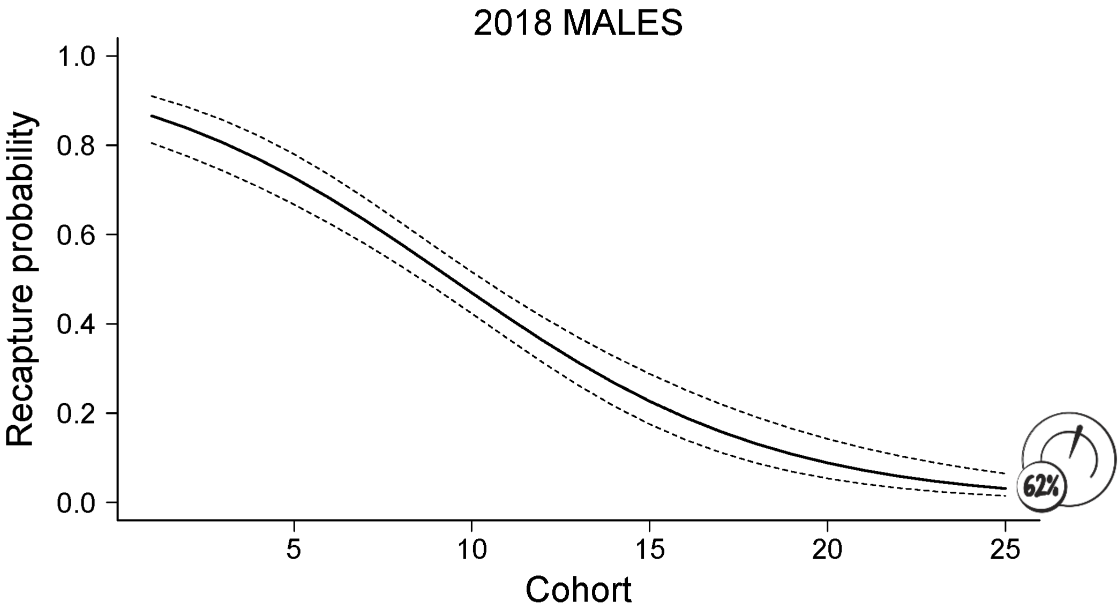


Figure 13: Estimated mean recapture probability for males (Phi(~DOF)p(~Cohort+relative humidity) model, median relative humidity in 2018 = 62%) in relation to the day of marking (Cohort) in 2018. Dashed lines represent 95% confidence intervals.

Males marked later had a strongly decreased recapture probability, whereas females marked later showed a slightly increased recapture probability, albeit only observed within a single year. Males marked later may have shown temporary emigration. It means they may have moved rather on the habitat edge and their daily range of movement only partially or occasionally overlapped with the study area.

We try to explain the difference between age, day of the flight period and cohort with Table 14. Individuals marked on the same day form a cohort (Table 14/c). Age is the days since first marking of the individual (Table 14/b), and day of the flight period are the number of days since the first Clouded Apollo was seen on wing on the study site (Table 14/a).

Table 14: Parameter Index Matrices (PIMs) for models including the day of the flight period (DOF) (a), Age (b), or Cohort (c). A PIM is a half-matrix representing parameter indices of a given model. Rows represent cohorts (groups of individuals marked on the same sampling

occasion) and columns represent sampling occasions or intervals. In models of the day of the flight period (a), all individuals have the same survival probability between two given sampling occasions and the same recapture probability on a given occasion, but parameter estimates vary during the flight season. In the age model (b), survival and recapture probabilities change with age in the same way for all individuals. For example, survival of individuals marked on the first day (cohort 1) between the first and second sampling occasions is the same as survival of individuals marked on the fourth day (cohort 4) between the fourth and fifth sampling occasions. For the cohort model (c), survival probabilities remain constant over time within cohorts but might differ among different cohorts. Note that, to facilitate the interpretation of models, these PIMs specify models where covariates are factorial, i.e. parameters Phi and p have 7 different values. In our model selection, we specified Age and Cohort models in a way that Phi and p were linearly dependent from age and cohort, respectively, thus only two parameters were estimated (intercept and slope).

a) Day of the flight period (DOF)

	sampling occasions						
	1	2	3	4	5	6	7
cohort 1.	**1**	**2**	**3**	**4**	**5**	**6**	**7**
cohort 2.		**2**	**3**	**4**	**5**	**6**	**7**
cohort 3.			**3**	**4**	**5**	**6**	**7**
cohort 4.				**4**	**5**	**6**	**7**
cohort 5.					**5**	**6**	**7**
cohort 6.						**6**	**7**
cohort 7.							**7**

b) Age

	sampling occasions						
	1	2	3	4	5	6	7
cohort 1.	**1**	**2**	**3**	**4**	**5**	**6**	**7**
cohort 2.		**1**	**2**	**3**	**4**	**5**	**6**
cohort 3.			**1**	**2**	**3**	**4**	**5**
cohort 4.				**1**	**2**	**3**	**4**
cohort 5.					**1**	**2**	**3**
cohort 6.						**1**	**2**
cohort 7.							**1**

c) Cohort

	sampling occasions						
	1	2	3	4	5	6	7
cohort 1.	**1**	**1**	**1**	**1**	**1**	**1**	**1**
cohort 2.		**2**	**2**	**2**	**2**	**2**	**2**
cohort 3.			**3**	**3**	**3**	**3**	**3**
cohort 4.				**4**	**4**	**4**	**4**
cohort 5.					**5**	**5**	**5**
cohort 6.						**6**	**6**
cohort 7.							**7**

5.2.4 Relationship between weather variables and apparent survival and between weather variables and recapture probability

Mean daily temperature was negatively related to apparent survival (Figure 14), meaning that individuals' survival probability was higher at lower temperatures. This effect was significant for females in 2016, 2017, 2018, 2019 and for males in 2014, 2016, 2017, 2018, 2019. In 2014 for males and 2018 for females, it was only significant together with the Time or Age variables. The only exception was in 2015 for females, when temperature had a positive effect on survival probability. Temperature was included in the most supported models in 2014 for males and in 2016 and 2019 for both sexes.

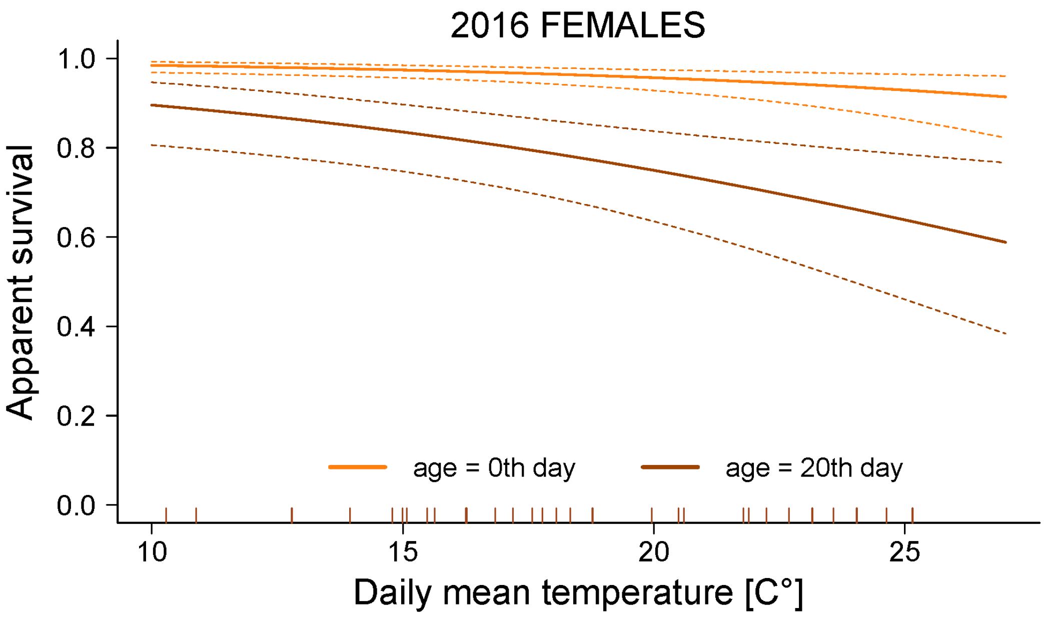


Figure 14: Estimated mean apparent survival for females (Phi(~Age+temperature)p(~Age+relative humidity) model, age = 0 day (light brown lines) and 20 (dark brown lines) days) in relation to the daily mean temperature in 2016. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the observed daily mean temperatures.

Daily mean temperature had a positive effect on recapture probability (Figure 15), i.e. when the temperature was higher, recapture probability was also higher. It was significant for females in 2014, 2015, 2018, 2019 and for males in 2014, 2015, 2016, 2018, 2019, but in the most supported model, it was included only in 2015 and 2019 for females. The only exception was in 2017 for males, when temperature had a negative effect on recapture probability and it was only significant together with the DOF and/or 'number of observers' variables.

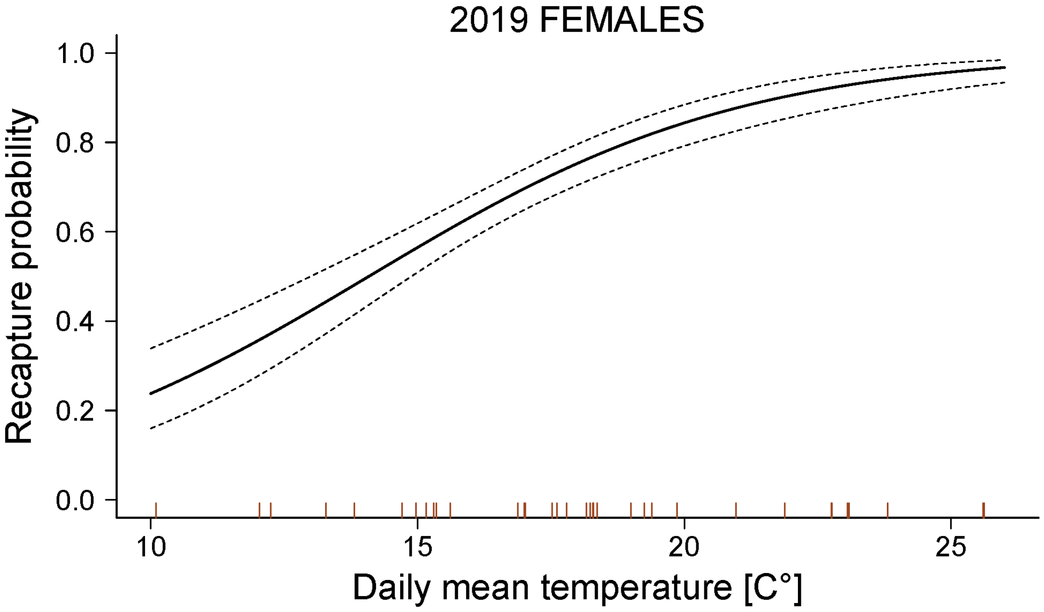


Figure 15: Estimated mean recapture probability for females (Phi(~Age+temperature+body mass)p(~temperature) model) in relation to the daily mean temperature in 2019. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the observed daily mean temperatures.

Mean daily relative humidity had a positive effect on apparent survival (Figure 16), which means that individuals' survival probability was higher at higher relative humidity. This relationship was significant for females in 2014, 2017, 2018, 2019 and for males in 2016, 2017, 2019. In 2014 and 2018 for females and in 2019 for both sexes it was only significant together with the Time or Age variables. The only exception was in 2015 for females, when relative humidity had a negative effect on survival probability. Mean relative humidity was included in the most supported models in 2014 and 2017 for females, and in 2017 for males.

By examining the most supported models and significant variables (Appendix 3), we see that temperature was a better predictor in some years, while relative humidity in others. In addition, there is a negative correlation between temperature and humidity, i.e. as temperature increases, humidity decreases and vice versa, therefore their effects cannot be actually separated.

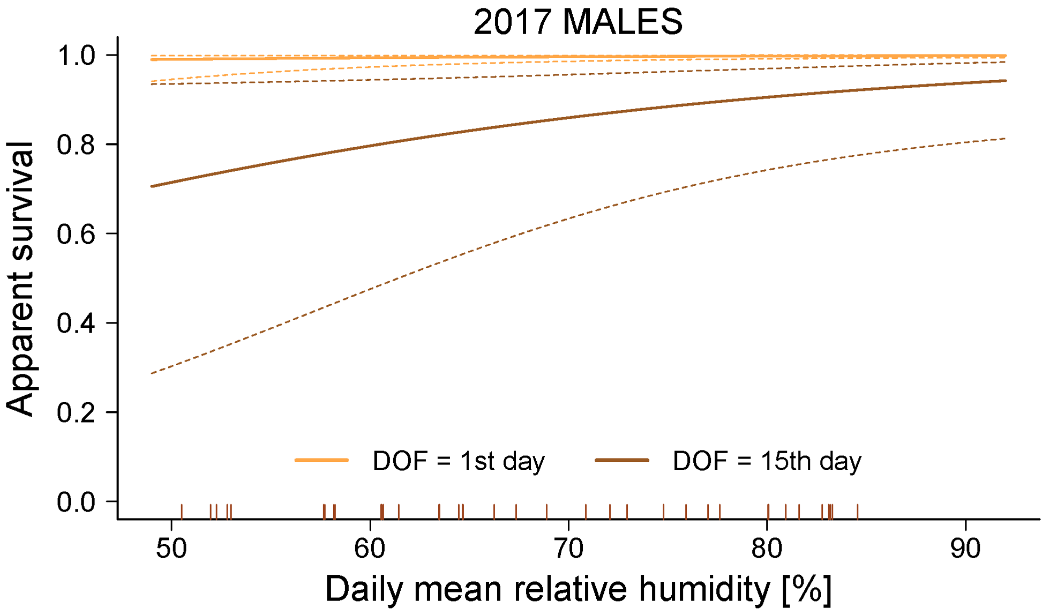


Figure 16: Estimated mean apparent survival for males (Phi(~DOF+relative humidity)p(~time) model, DOF (Day of the flight period) = 1st and 15th day) in relation to the daily mean relative humidity in 2017. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the observed daily mean relative humidity.

Daily mean relative humidity had a negative effect on recapture probability (Figure 17), so when relative humidity was lower, recapture probability was higher. It was significant for females in 2014, 2015, 2016, 2018, 2019 and for males in 2014, 2016, 2017, 2018, 2019, but in the most supported model, it was included only in 2014 and 2016 for females, and in 2018 for males.

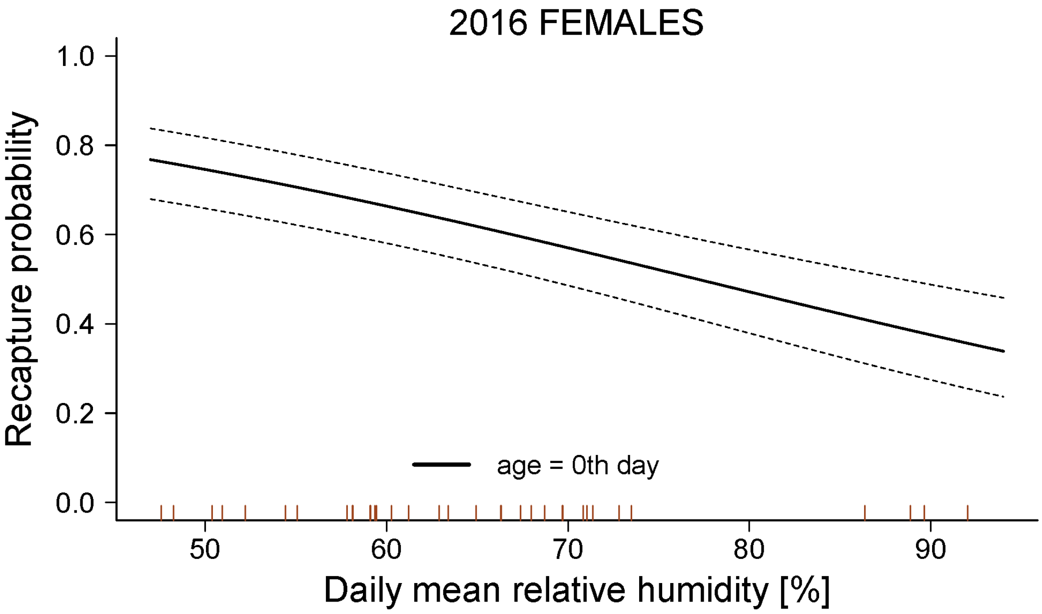


Figure 17: Estimated mean recapture probability for females (Phi(~Age+temperature)p(~Age+relative humidity) model, age = 0th day) in relation to the daily mean relative humidity in 2016. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the observed daily mean relative humidity.

We observed that as age increased, the effect of temperature was greater on apparent survival (Figure 14). High daily temperature can cause unfavourable physiological effects in insects (Chown & Terblanche, 2006) including membrane and protein structure disruption (Hochachka & Somero, 2002), alterations in the cellular microenvironment, DNA damage (Feder, 1999; Somero, 1995) or elevated metabolic rates (González-Tokman et al., 2020), accelerating their ageing process, potentially leading to a decline in survival (Niitepõld, 2010). Moreover, elevated temperatures may reduce immune response of butterflies (Karl et al., 2011) and extreme heat can seriously reduce their survival (Klockmann et al., 2016).

Our results showed that at the beginning of the flight period relative humidity had a weak effect, but towards the end of the season it had a stronger effect on apparent survival (Figure 16). We suspect that in the second half of the season smaller individuals emerged and smaller individuals may be more adversely affected by the weather (see section 5.1.4., and Pásztor *et al.*, 2022). At lower relative humidity, in drier conditions, temperature is higher, water loss increases due to intensified evaporation which may have detrimental effects on butterflies (Mazer & Appel, 2001), potentially leading to desiccation stress and increased mortality. Hot and/or dry conditions were typical at the end of the flight periods, likely contributing to the declining survival probability with the progress of the flight period, while potentially playing a role in the higher recapture probability due to decreased activity.

Vlašánek and colleagues observed increased survival probability and decreased recapture probability with rising temperature and precipitation, particularly in male Clouded Apollos (Vlašánek et al., 2009). This is contrary to our results, except in 2015 when we found the same for female survival increasing with rising temperature. The phenomenon observed by Vlašánek may be attributed to males patrolling even under suboptimal conditions. Furthermore, their study field is geographically much farther north (Southeast Czech Republic, 48°49′N, 16°42′E) than ours (North-Central Hungary, 47°45'22.7"N, 19°2'53.4"E); thus, the mean daily temperature was likely lower than ours throughout the flight period (no exact temperature data given in the article), creating more optimal conditions for butterflies' activity. In 2015, within our study field, the mean temperature for this flight period was 18.77°C, which is lower than in 2018 and 2019 but higher than in 2014, 2016, and 2017 (Table 11, Appendix 2Appendix 7). We observed the highest maximum mean daily temperature in 2015 at 26.7°C. Importantly, this temperature peak occurred in the second half of the flight period, and following that day, we noted a sustained drop in temperature to below 20°C over the next eight days (Appendix 2Appendix 7). This temperature pattern was unique among flight periods in other years. We propose that the impact of temperature on Clouded Apollo survival varies across different temperature ranges.

5.2.5 Relationship between body sizes and apparent survival, and between body sizes and recapture probability

Initial body mass had a significant positive effect on apparent survival in 2019 for females (Figure 18), and it was included in the most supported model in this year. For males in 2019, it also had a significant positive effect but only together with Age and/or the mean daily temperature.

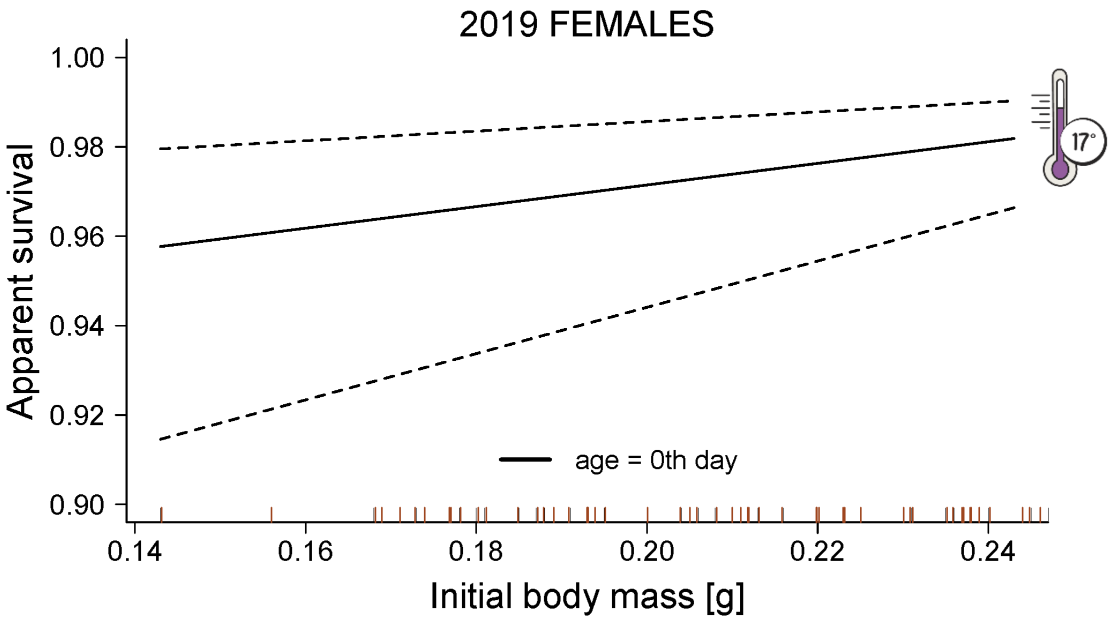


Figure 18: Estimated mean apparent survival for females (Phi(~Age+temperature+body mass)p(~temperature) model, age = 0th day, daily median temperature = 17 °C) in relation to the initial body mass in 2019. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the initial body mass of recaptured individuals.

In case of recapture probability, the direction of the effect of initial body mass differed from year to year. It had a significant positive effect in 2014 and 2019 for males, whereas it had a negative effect in 2018 for males in which case the recapture probability was much lower. Furthermore, body mass was only significant together with mean daily temperature or relative humidity in 2016, exhibiting a positive effect and in 2019, showing a negative effect for females.

Initial body mass was not included in the most supported models as a covariate of recapture probability in any of the years.

Initial thorax width had a significant positive relationship with apparent survival in 2015 (Figure 19) and in 2018 for both sexes. It had a significant positive effect only together with Age and/or mean daily temperature or relative humidity or DOF in 2014, 2019 for both sexes. It was included in the most supported model only in 2015 for males, but it was significant only together with DOF. Positive effect means that individuals with wider thoraces had higher survival probabilities.

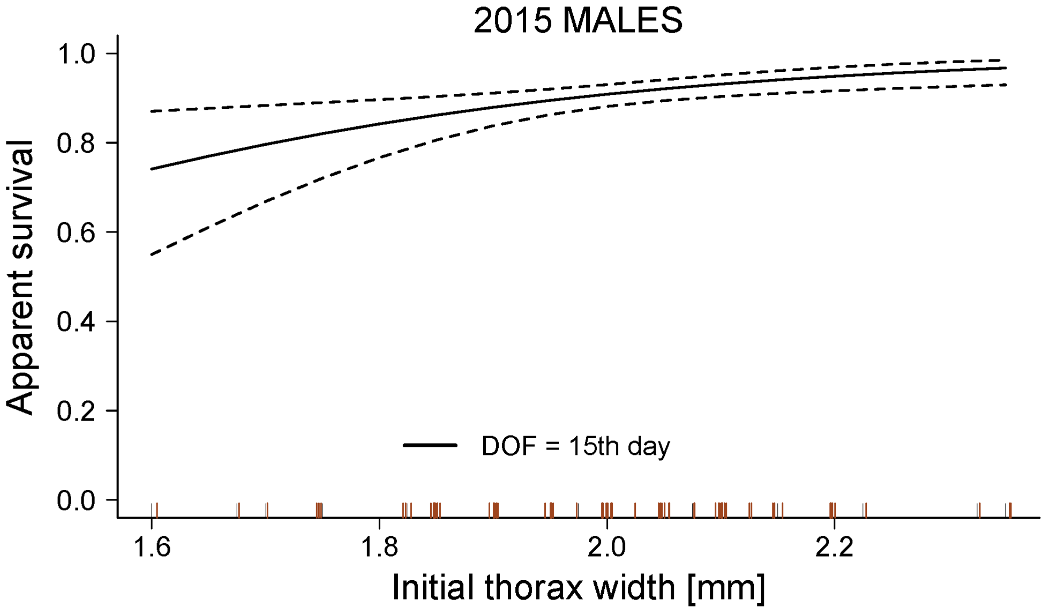


Figure 19: Estimated mean apparent survival for males (Phi(~DOF+thorax)p(~observers+Cohort+wing+wing^2) model, DOF = 15th day (max = 27 days)) in relation to the initial thorax width in 2015. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the initial thorax width of recaptured individuals.

In case of recapture probability, the direction of the effect of thorax width differed from year to year. It had a significant positive effect in 2014 and 2015 for females (Figure 20) and in

2018 and 2019 for males, whereas it had a negative effect in 2014 and 2015 for males. Furthermore, thorax width was only significant together with the number of observers in 2014 and 2019 for males. In the most supported models, thorax width was included only for females in 2014 and 2015.

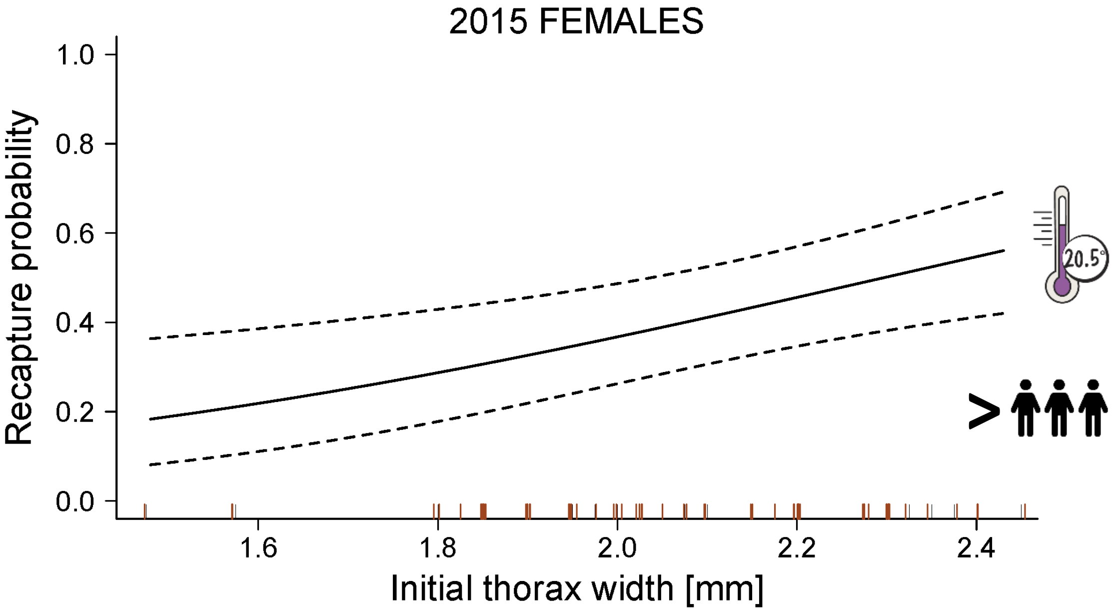


Figure 20: Estimated mean recapture probability for females (Phi(~DOF)p(~observers+temperature+thorax) model, median temperature = 20.5 °C, observers > 3 person) in relation to the initial thorax width in 2015. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate the initial thorax width of recaptured individuals.

Wing length had a significant positive effect on apparent survival in 2017 and 2019 for males (Figure 21), meaning that individuals with longer wings had higher survival probabilities. The most supported model contained wing length only in 2019, and in 2017 wing length was only significant when the model included Age as well. However, the interaction between Age and wing length was not significant in any year.

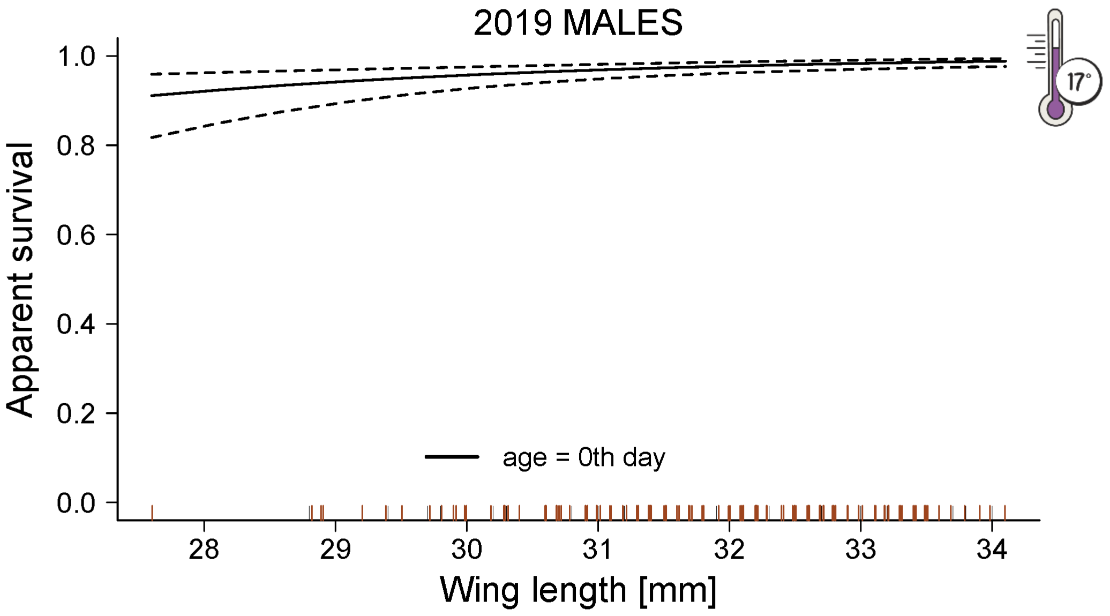


Figure 21: Estimated mean apparent survival for males (Phi(~Age+temperature+wing)p(~time) model, age = 0th day, daily median temperature = 17 °C) in relation to wing length in 2019. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate wing length of recaptured individuals.

Wing length had a significant negative effect on recapture probability (Figure 22) in 2015 for both sexes and in 2019 for females, but the most supported model contained wing length and its quadratic term only in 2015 for males. It means that recapture probability was lower for individuals with longer wings. In 2018 for males, wing length had a significant positive effect on recapture probability but only when the model contained daily mean temperature or daily mean relative humidity and/or Time too.

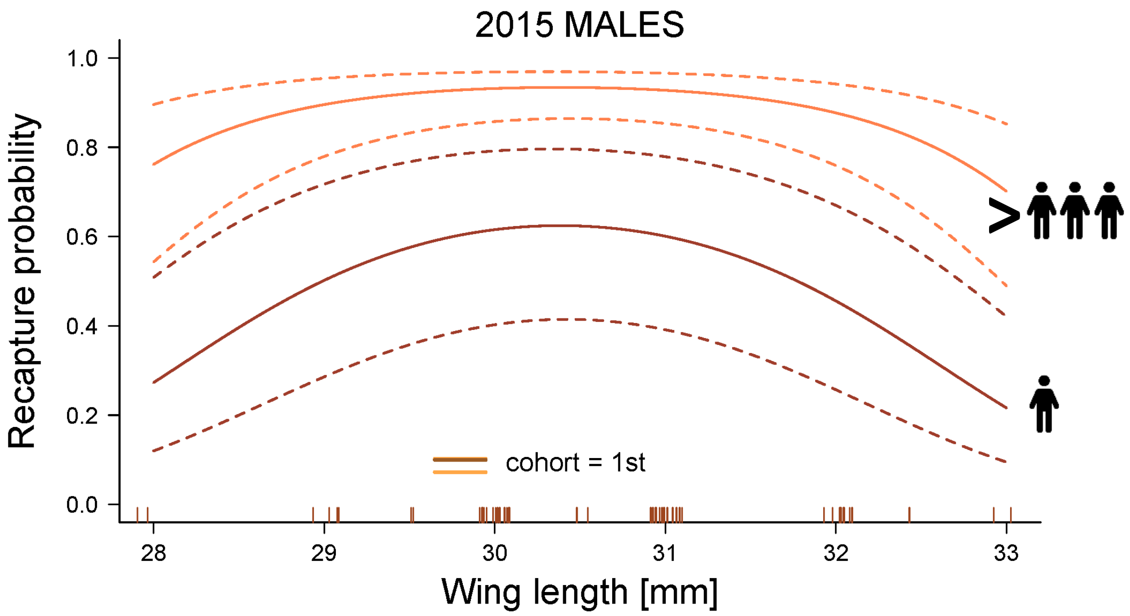


Figure 22: Estimated mean recapture probability for males (Phi(~DOF+thorax)p(~observers+Cohort+wing+wing^2) model, number of observers > 3 (light brown) or 1 (dark brown), cohort = 1st (for both light and dark brown) (max = 31)) in relation to the wing length in 2015. Dashed lines represent 95% confidence intervals. Rugs on the x-axis indicate wing length of recaptured individuals.

Proboscis length had a significant positive effect on survival probability for males in 2016 and 2017, and for both sexes in 2019, but only together with Age and/or daily mean temperature or relative humidity. Positive effect means that apparent survival was higher in individuals with longer proboscides. Proboscis length was not included in the most supported models in any of the years, and the interaction between Age and proboscis length was not significant either.

Proboscis length had a significant negative effect on recapture probability for females in 2019, but only together with daily mean relative humidity. This means that recapture probability was lower in butterflies with longer proboscides. It was not included in the most supported models in any of the years.

Resource availability during larval and adult stages significantly influences survival and fitness in Lepidoptera. Inadequate resources during these stages often lead to decreased survival rates and compromised fitness in adults (Bauerfeind et al., 2009; Bauerfeind & Fischer, 2009; C. L. Boggs & Freeman, 2005). We found that larger Clouded Apollo individuals had better survival, especially thorax width and wing length showed a strong positive relationship with survival. Larger body size is likely to be related to the quality and quantity of larval food, implying that the availability and quality of the food source may indirectly influence Apollo survival.

Larger body mass and wider thoraces may signify larger reserves available for somatic maintenance, potentially leading to higher survival probabilities. Larger body sizes tend to contain increased energy reserves, which also can serve as a buffer against environmental stressors and resource fluctuations, ultimately contributing to improved survival rates. This link between larger body mass and enhanced survival probability highlights the significance of body size as a crucial determinant of resource allocation strategies impacting butterfly fitness and survival. In line with this, we revealed phenotypic senescence, body size decline with age, in the study population of Clouded Apollos (See section 5.1.2).

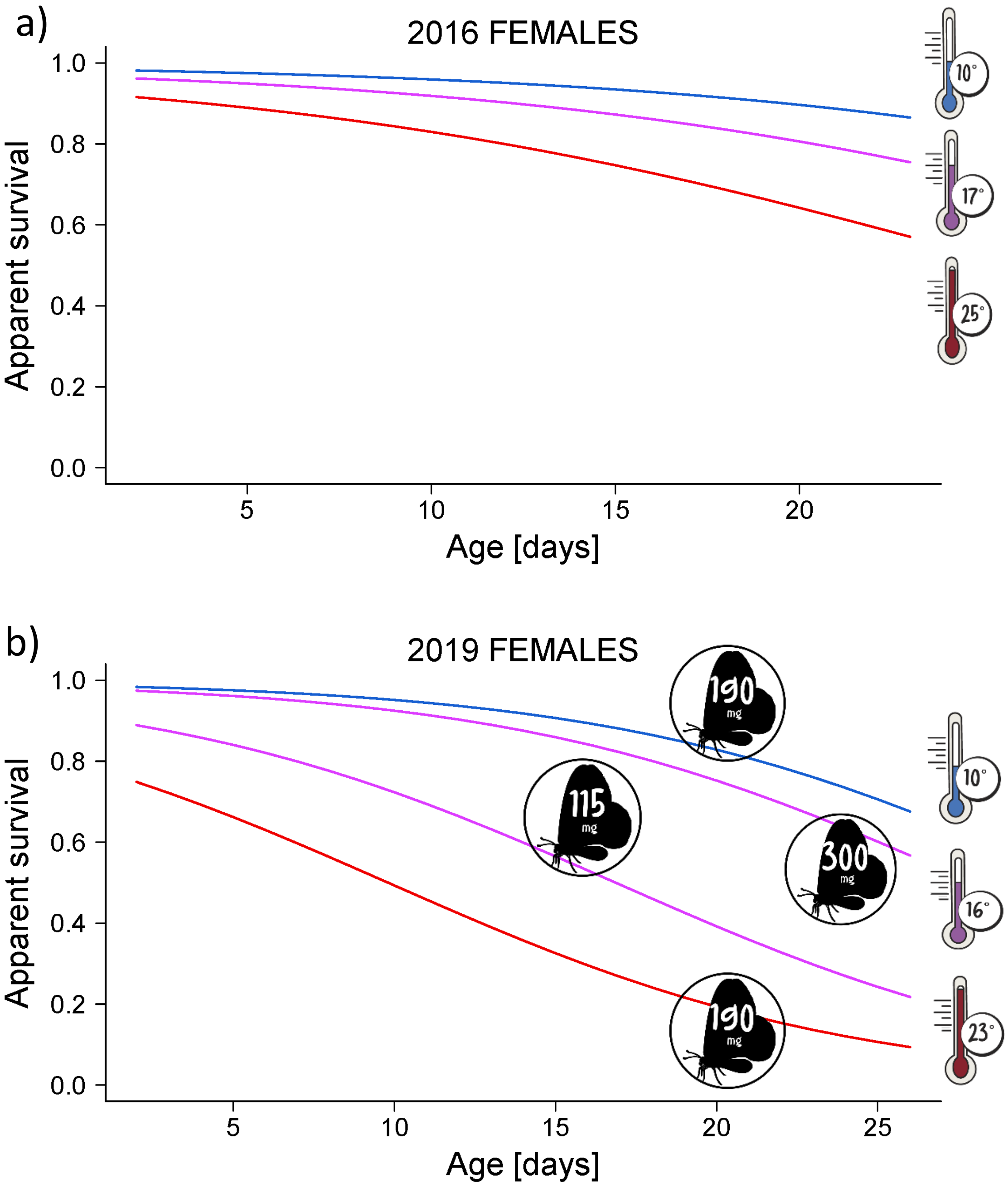
Wider thoraces and longer wings may indicate better flight ability (Altizer & Davis, 2009; Davis & Holden, 2015; Sekar, 2012; Soule et al., 2020), which allows individuals to be more active, flying longer distances and flying above the canopy (often observed in Clouded Apollos during field work) resulting in a lower recapture probability.

We note that body size parameters did not appear in the most supported models in every study year and their effects on survival and recapture probability were rather weak in some cases.

5.2.6 The combined effects of different variables on apparent survival and recapture probability

5.2.6.1 Apparent survival

Butterflies' age had a negative effect on apparent survival, but the degree of the effect differed according to daily mean temperature and/or individuals' body sizes (initial body mass, wing length). Survival probability was higher at lower temperatures (Figure 23/a-c, blue solid line) and with larger body sizes (Figure 23/b, c, pink lines). At the same temperature, individuals with smaller body sizes (Figure 23/b, c, pink lines) had lower survival probabilities. These figures can be interpreted as butterflies aged faster at higher temperatures or as temperature had a stronger negative effect on survival at older age. Similarly, smaller butterflies aged faster or body size had a stronger positive effect on survival at older age.



c)

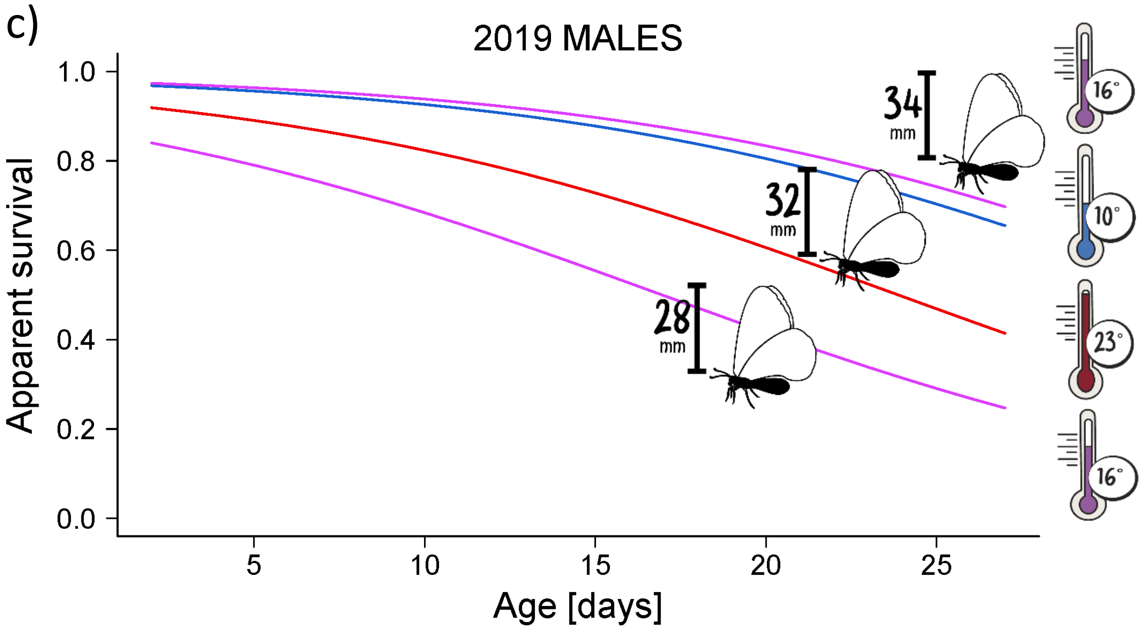
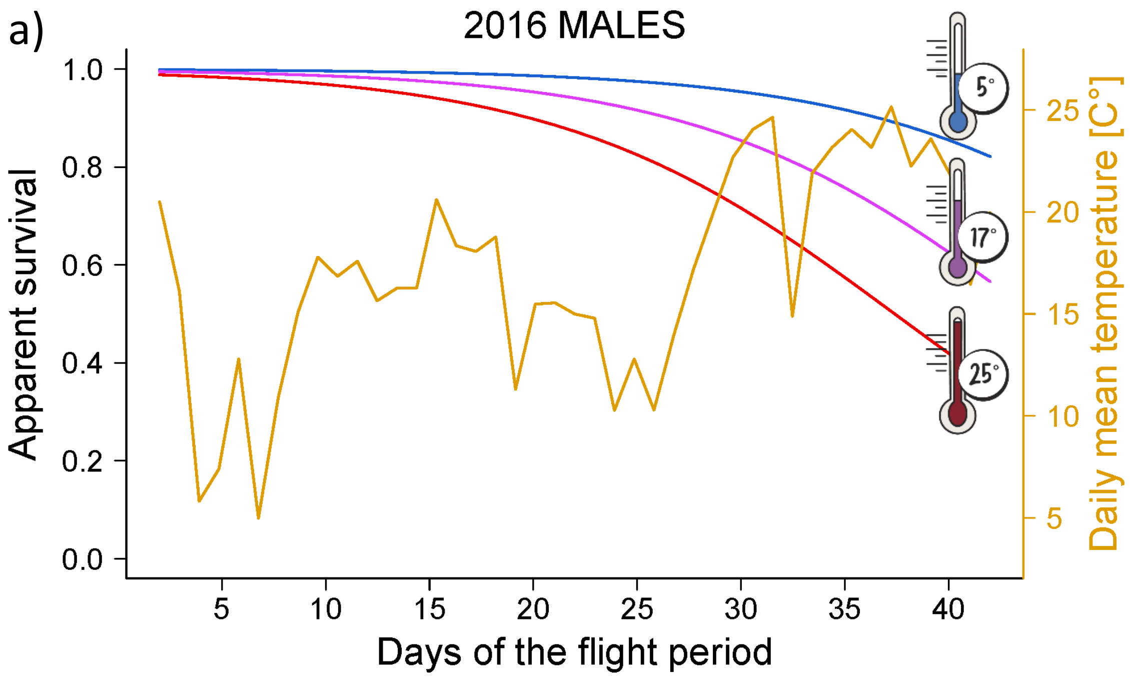


Figure 23: Estimated mean apparent survival in relation to the number of days since marking (Age) and a) minimum (blue line), median (pink line) and maximum (red line) temperature for females in 2016 (Phi(~Age+temperature)p(~Age+relative humidity) model), or b) minimum (blue line), median (pink line) and maximum (red line) temperature and minimum (pink line), median (red and blue lines) and maximum (pink line) initial body mass for females in 2019 (Phi(~Age+ temperature+body mass)p(~temperature) model), or c) minimum (blue line), median (pink line) and maximum (red line) temperature and minimum (pink line), median (red and blue lines) and maximum (pink line) wing length for males in 2019 (Phi(~Age+temperature+wing)p(~time) model).

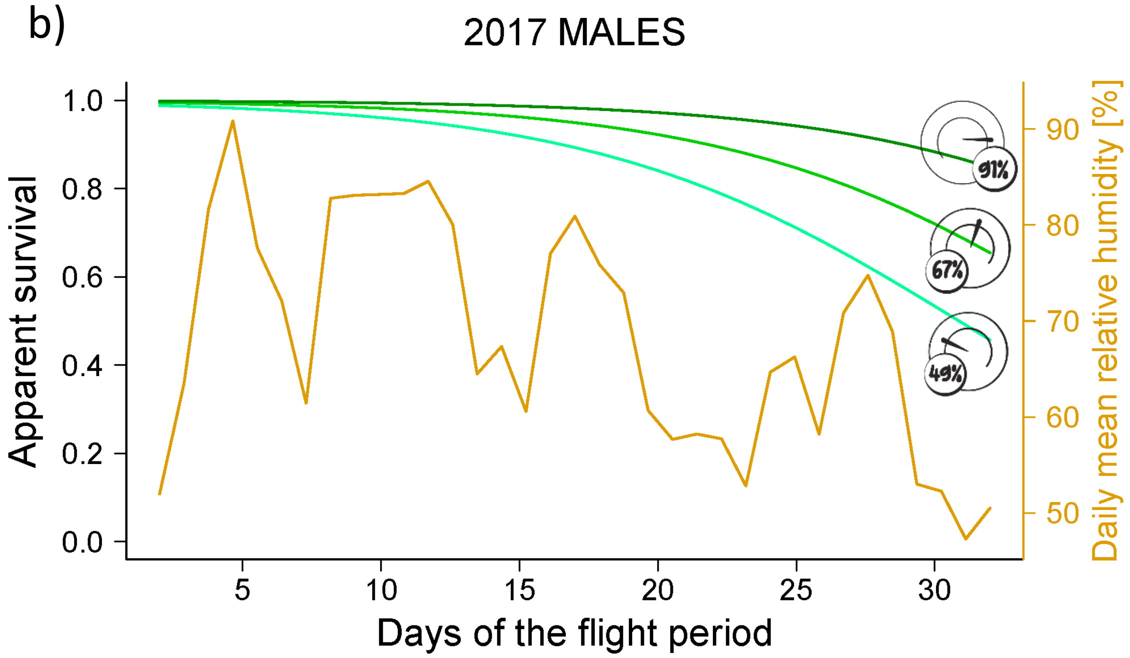
The day of the flight period was negatively related to apparent survival, but the degree of the effect differed according to the daily mean temperature or relative humidity and/or individuals' body sizes (initial thorax width). Simultaneously, survival probability was higher at a lower temperature (Figure 24/a, blue line), at a higher relative humidity (Figure 24/b, the darkest green line) and with larger thorax width (Figure 24/c, the darkest purple line) changing with the progress of the flight period.

a)



2016 MALES

b)



2017 MALES

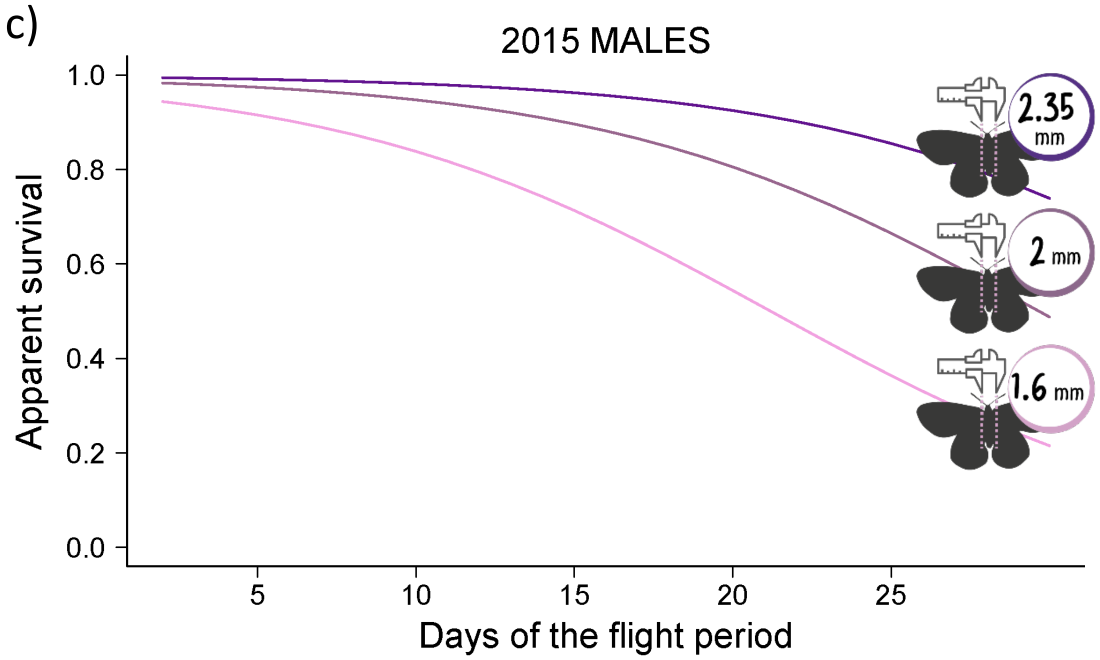


Figure 24: Estimated mean apparent survival in relation to the day of the flight period (DOF) and a) minimum (blue line), median (pink line) and maximum (red line) temperature for males in 2016 (Phi(~DOF+temperature)p(~time) model), or b) minimum (light green line), median (medium green line) and maximum (dark green line) relative humidity (RH) for males in 2017 (Phi(~DOF+relative humidity) p(~time) model), or c) minimum (light violet line), median (medium violet line) and maximum (dark violet line) thorax width for males in 2015 (Phi(~DOF+thorax) p(~observers+Cohort+wing+wing^2) model). Yellow solid lines represent a) the daily mean temperature in the shaded areas of the study site or b) the daily relative humidity in the shaded areas of the study site along the days of the flight period.

The relationship between body sizes, weather variables, and survival probability in butterflies unveils a complex interplay influenced by environmental conditions. Environmental conditions, such as temperature and humidity, have a significant impact on insect behaviour and physiology (Heinrich, 2013). Furthermore, weather variables such as temperature, humidity, and precipitation play key roles in shaping butterfly survival probabilities (Kingsolver et al., 2012). Studies detected higher survival probability at lower daytime temperature (Bauerfeind et al., 2009; Klockmann et al., 2016), but reduced lifespan in male butterflies at higher night temperature (Rosa & Saastamoinen, 2021). Karl and his colleagues observed increased body mass and abdominal fat levels with increasing temperature, suggesting that this phenomenon may be linked to higher (feeding) activity (Karl et al., 2011).

Temporal shift in the age structure of the population (i.e. increasing mean age with the progress of the flight period) aligns with higher mean daily temperatures in the latter half of the season. As ambient temperature (up to ca. 30 °C) is positively related to butterfly activity (Casula & Nichols, 2003), this might also contribute indirectly to higher recapture probability later in the flight season.

5.2.6.2 Recapture probability

We note that in most years the most supported model contained a full-factorial time-dependent recapture probability, which means that for each sampling occasion a different parameter value was estimated. In these cases, there must have been such a high variation in recapture probability that the covariates could not explain it properly. However, some covariates could have a significant effect in these datasets as well if we compared them to the constant model.

Recapture probability was higher in older butterflies, and at lower relative humidity (Figure 25, the lightest green line). Recapture was higher at lower humidity for individuals of the same age.

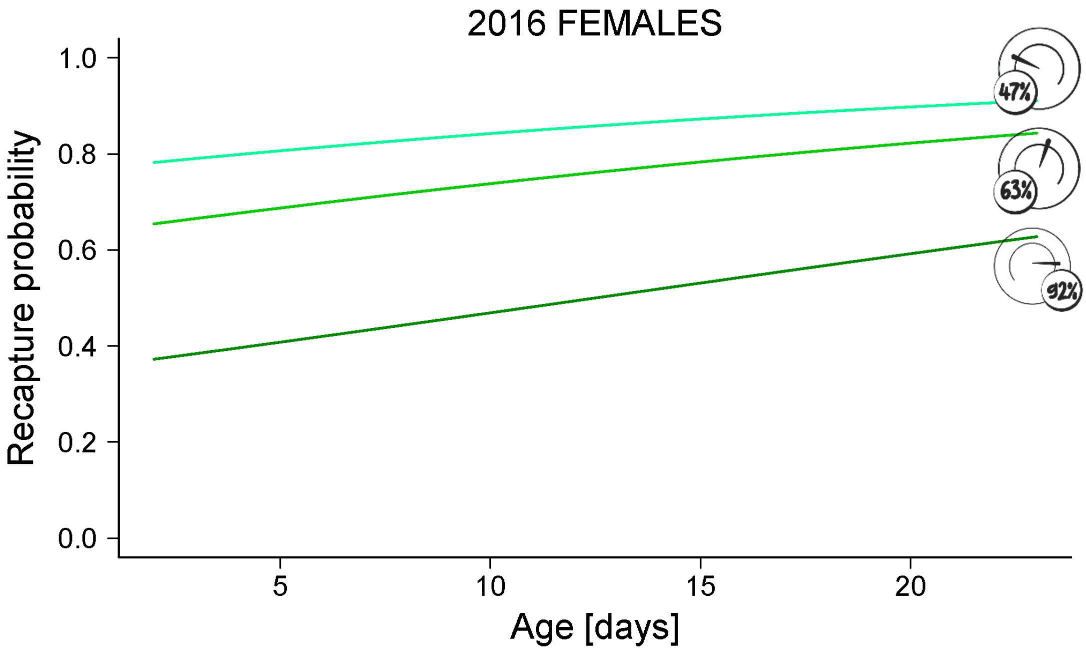


Figure 25: Estimated mean recapture probability in relation to the number of days since marking (Age) and minimum (light green line), median (medium green line) and maxium (dark green line) relative humidity (RH) for females in 2016 (Phi(~Age+temperature)p(~Age+relative humidity) model).

Daily mean temperature, the number of observers and initial thorax width all had a positive effect on recapture probability, meaning that it was higher at higher temperature, when more observers (>3 people) worked in the field (Figure 26, dashed line) and in case of individuals with wider thoraces (Figure 26, the darkest purple dashed and solid lines). When the number of observers was the same, recapture was better for butterflies with wider thoraces.

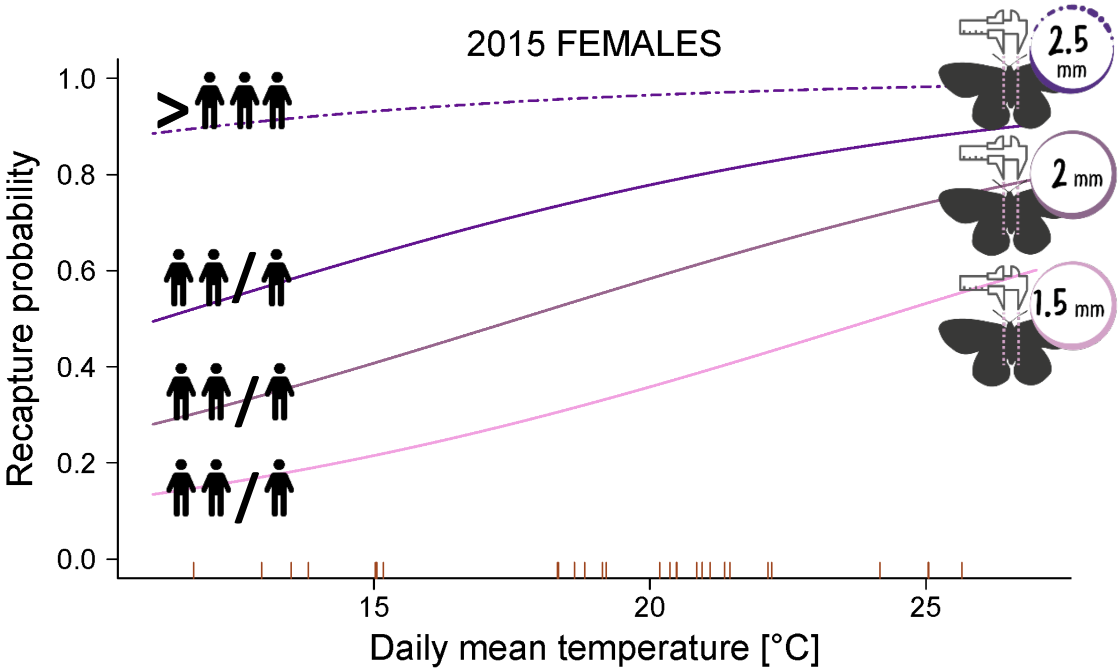


Figure 26: Estimated mean recapture probability in relation to the daily mean temperature in the shaded areas of the study site, number of observers (2–3 people or more than 3 people) and minimum (light violet line), median (medium violet line) and maximum (dark violet solid and dashed lines) thorax width for females in 2015 (Phi(~DOF)p(~observers+temperature+thorax) model). Rugs on the x-axis indicate the daily mean temperatures.

Daily mean relative humidity had a negative effect on recapture probability, which means that it was higher at lower humidity. Simultaneously, recapture was better for butterflies with wider thoraces (Figure 27, the darkest purple line).

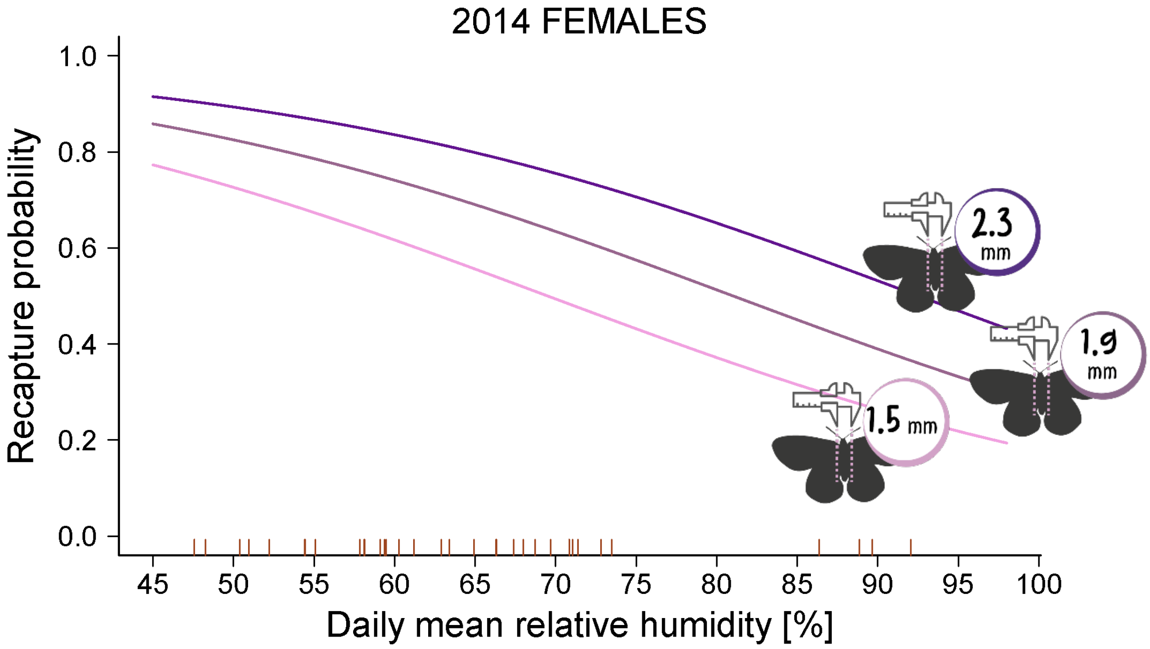


Figure 27: Estimated mean recapture probability in relation to the daily mean relative humidity in the shaded areas of the study site and minimum (light violet line), median (medium violet line) and maximum (dark violet line) thorax width for females in 2014 (Phi(~DOF+relative humidity)p(~relative humidity+thorax) model). Rugs on the x-axis indicate the daily mean relative humidity.

Higher temperatures are known to elevate insect activity levels, influencing their flight patterns and overall movement (Heinrich, 2013; Kingsolver et al., 2012). Evidently, when more observers work on the field, more observations are performed, especially on larger butterflies.

6. CONCLUSIONS AND RECOMMENDATIONS

We conducted a long-term (7 consecutive years) study in a natural population of Clouded Apollo butterflies using mark-recapture method complemented with continuous recording of weather parameters and precise measurements of body size of individual butterflies.

We found a significant decrease in body mass and thorax width with age (phenotypic senescence) in both sexes in each year (2014–2020). We observed large variation in initial body size and smaller variation in the rate of senescence (body mass and thorax width decline with age) among individuals. Females had larger initial body sizes but they declined faster compared to males, indicating that differential selection influences phenotypic senescence.

We also proved that apparent survival declined with age (actuarial senescence) in this Clouded Apollo population in almost every year (2014–2017 and 2019). The most supported models of CJS analyses were not the same for all years, but we found that the direction of the relationships was the same in almost every case. Apparent survival also declined with the day of the flight period in all years (2014–2019) and both sexes. In most years, survival was negatively related to temperature and positively to relative humidity, in some other years body size was positively related to survival, which means that larger individuals had a higher survival probability. The relationship between body size and survival probability was less clear and weaker compared to that with age and weather parameters.

Recapture probability, in some years, was significantly higher when more observers worked in the field, when temperature was higher and relative humidity was lower. Furthermore, in some other years, recapture probability increased with age and body size, i.e. older and/or larger individuals were recaptured with higher probability.

To our knowledge, this is the first study that revealed phenotypic senescence in a natural butterfly population, using *in situ* non-invasive measurements. Furthermore, we were able to detect actuarial senescence and revealed how body size and weather variables affected Clouded Apollo butterflies' survival and recapture probability.

Our results suggest that the rates of senescence and larval growth may be influenced by individually and annually varying environmental variables and genetic factors. Body size largely determines fecundity (Honěk, 1993; Tammaru et al., 2002) and senescence may also affect reproductive success in insects (Boggs, 1997; Wickman & Karlsson, 1987; Wiklund & Karlsson, 1984). Therefore, we recommend that laboratory studies in the future should aim to (i) uncover the separate effects of certain weather conditions (temperature, humidity etc.) and resource availability on the rates of larval growth, adult survival and senescence, (ii) investigate the genetic background and its relationship with environmental factors of ageing of insects. In addition, more long-term field studies in natural populations of butterflies should be conducted in order to reveal the combined effects of different environmental factors on the survival probability of individuals. Ultimately, a deeper understanding of these relationships would help us better predict the effects of current global environmental changes on the viability of insect populations.

If we knew the vulnerabilities of these butterflies, it would be easier to take appropriate steps to mitigate the effects of climate change and habitat loss, e.g. with appropriate habitat management. For Clouded Apollos, the most important aspect is the patchiness of the habitat, because this heliophilous butterfly needs open sunny spots for basking, but interspersed with shrubs for resting in the shadow to avoid overheating in hotter days (Konvička & Kuras, 1999). Other important factors are the wide variety of dense herbaceous plants as nectar source for feeding, and the transition zone between the meadow and the forest, where they lay their eggs near to *Corydalis* species, their larval food plants (Habel et al., 2022). Clouded Apollo butterflies seem to be an appropriate species for in-situ measurements and mark-recapture studies because of their size, behaviour and their robust body structure.

Understanding the interplay between survival, life history traits, and fitness is essential for comprehending the adaptive strategies of Lepidoptera in response to environmental pressures and for predicting their population dynamics in changing ecosystems. In order to detect trends and control for intra- and interspecific longevity and ageing (Van de Pol & Verhulst, 2006), longitudinal research in consecutive generations is necessary (Nussey et al., 2008). These scientific results can later be applied to conservation biology research and management plans for species conservation programmes. Future research could play a major role in determining the maximum temperature that the species can tolerate, which, combined with local climate predictions, could help to select habitats where the studied butterfly population can be protected and sustained in the long term.

7. NEW SCIENTIFIC RESULTS

1) This is the first study where body mass and thorax width were repeatedly measured on butterflies during their lifespan *in situ* non-invasive in their natural habitat.
2) We revealed phenotypic senescence in a natural population of Clouded Apollo butterflies. We detected that both body mass and thorax width decreased with age, proving that physiological ageing exists in natural circumstances in butterflies. We found sexual differences in the rate of phenotypic senescence, but despite the annual variation of initial body sizes, the rate of senescence did not vary considerably across years.
3) We provided evidence that actuarial senescence occurred almost every year (2014–2017 and 2019) in this natural population of Clouded Apollos, which means that the survival probability of butterflies decreased with age.

4) We found that ambient temperature and relative humidity significantly influenced butterfly survival. Butterflies had higher survival at cooler temperature and higher relative humidity.

5) We also found that body size, especially thorax width and wing length, was positively related to butterfly survival in some years, indicating that larger butterflies survive better.

6) The effects of weather variables and body size were more apparent at an older age of Clouded Apollos.

7) In some years, detectability of butterflies was significantly affected by weather parameters and body size. The recapture probability was higher at higher temperatures and lower relative humidity, as well as in larger butterflies.

8. SUMMARY

To deepen our knowledge on the dynamics of butterfly populations, it is needed to reveal how environmental variables and body sizes are related to survival, senescence and detectability of butterflies. Senescence seems to be universal in living organisms and plays a major role in life history strategies. Phenotypic senescence, the decline of body condition and/or performance with age, is a largely understudied component of senescence in natural insect populations. Actuarial senescence, the decline in survival probability with age, is not a long-studied area, only observed in the early 2000s to exist in natural insect populations (Bonduriansky & Brassil, 2002). However, it would be important to understand how and why insects age under natural conditions.

We aimed to investigate how body mass and thorax width change with age in the Clouded Apollo butterfly (*Parnassius mnemosyne*) and to assess the relationship of this change with sex and wing length. Our further goal was to uncover the effects of different weather parameters and body sizes on survival and recapture probability for this butterfly species.

We studied a single, natural population of the univoltine Clouded Apollo butterfly between 2014 and 2020 using mark-recapture during the whole flight period each year. Repeated *in situ* measurements of body mass and thorax width and single measurements of wing and proboscis length were performed on marked individuals. We analysed body mass and thorax width change with age (days since marking), wing length and the date of the first capture. We also measured temperature and relative humidity with data loggers. Data were analysed using Linear Mixed Effects models and the Cormack-Jolly-Seber model.

Both body mass and thorax width declined non-linearly with age. Individuals appearing earlier in the flight period had significantly higher initial body mass and thorax width and their body mass declined faster than later ones. Initial body sizes of females were higher, but males' body sizes decreased slower. Initial thorax width showed higher annual variation than body mass.

In all years, butterfly survival declined with the progress of the flight period or age in both sexes. Generally, at lower temperature, higher relative humidity and with larger body size,

individuals had higher survival probabilities. Recapture probability increased with temperature, number of observers, body size and age, and it declined with relative humidity. However, these relationships were significant only in a few years. We found relatively small differences between the sexes, but a high annual variation in the ranking of models.

To our best knowledge, this is the first study that revealed phenotypic senescence in a natural butterfly population, using *in situ* non-invasive measurements. We found sexual differences in the rate of phenotypic senescence. Despite the annual variation of initial body sizes, the rate of senescence did not vary considerably across the years.

Our results provide a deeper insight into the demography of butterfly populations and may enable us to reveal potential pathways of adaptation to environmental changes.

9. ÖSSZEFOGLALÓ

A lepkepopulációk dinamikájára vonatkozó ismereteink elmélyítése érdekében fel kell tárni, hogy a környezeti változók és a testméretek hogyan kapcsolódnak a lepkék túléléséhez, öregedéséhez és észlelhetőségéhez. Úgy tűnik, hogy az öregedés univerzális jelenség az élő szervezetekben, és fontos szerepet játszik az életmenet stratégiákban. A fenotípusos öregedés, azaz a test állapotának romlása és/vagy a teljesítmény csökkenése az életkor előrehaladtával, vizsgálatáról természetes rovarpopulációkban nem tudunk. Csak a 2000-es évek elején figyelték meg, hogy a túlélési valószínűség csökkenése a korral egyáltalán létezik természetes rovarpopulációkban (Bonduriansky & Brassil, 2002), de az azóta eltelt két évtizedben alig néhány vizsgálat irányult erre. Pedig fontos lenne megérteni, hogyan és miért öregednek a rovarok természetes körülmények között, hogy jobban előre jelezhessük a populációk és a fajok válaszát napjaink környezeti változásaira.

Célunk annak vizsgálata volt, hogy hogyan változik a testtömeg és a tor szélessége a kis Apolló-lepkék életkorával, és ez a változás milyen kapcsolatban áll az ivarral és a szárnyhosszal. További célunk volt, hogy feltárjuk a különböző időjárási paraméterek és testméretek hatását a túlélésre és a visszafogási valószínűségre ennél a lepkefajnál.

2014 és 2020 között jelölés-visszafogás módszerrel vizsgáltunk egy természetes kis Apolló-lepke populációt. A megjelölt egyedeken a testtömeg és a torszélesség ismételt *in situ* mérését, valamint a szárny és a pödörnyelv hosszának egyszeri mérését végeztük el. Elemeztük a testtömeg és a tor változását a kor (a jelölés óta eltelt napok száma), a szárnyhossz és az első befogás időpontja függvényében. A hőmérsékletet és a relatív páratartalmat is mértük a kutatási helyszínre egész évben kihelyezett mérő műszerekkel. Később ezen változók kapcsolatát lineáris kevert modellekkel és Cormack-Jolly-Seber-modellekkel elemeztük.

Mind a testtömeg, mind a tor szélessége nem lineárisan csökkent az életkorral. A repülési időszakban korábban megjelenő egyedek kezdeti testtömege és torszélessége szignifikánsan nagyobb volt, és testtömegük gyorsabban csökkent, mint a később megjelenteké. A nőstények kezdeti testmérete nagyobb volt, de a hímek testmérete lassabban csökkent. A tor kezdeti szélessége nagyobb éves ingadozást mutatott, mint a testtömeg.

A lepkék túlélése minden évben csökkent a repülési időszak vagy az életkor előrehaladtával mindkét ivarnál. Általában alacsonyabb hőmérsékleten, magasabb relatív páratartalom mellett és nagyobb testméret esetén az egyedek túlélési valószínűsége magasabb volt. A visszafogás valószínűsége a hőmérséklettel, a megfigyelők számával, a testmérettel és az életkorral nőtt, a relatív páratartalommal pedig csökkent. Ezek az összefüggések azonban csak néhány évben voltak szignifikánsak. Az ivarok között viszonylag kis különbségeket találtunk, de a legjobb modellek évenként eltértek.

Legjobb tudomásunk szerint ez az első olyan tanulmány, amely a fenotípusos öregedést természetes lepkepopulációban mutatta ki, *in situ* nem invazív mérések felhasználásával. A nőstények testmérete szignifikánsan gyorsabban csökkent az életkorral, mint a hímeké. A kezdeti testméretek éves változása ellenére az öregedés mértéke nem változott jelentősen az évek között. Eredményeink mélyebb betekintést nyújtanak a lepkepopulációk demográfiájába, és támogatják a környezeti változásokhoz való alkalmazkodás lehetséges útjainak feltárását.

10. APPENDICES

Appendix 1: Bibliography

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Appendix 2: Mean daily temperature and relative humidity during flight periods for all years.

Light blue = minimum temperature for the given year, Dark red = maximum temperature for the given year,
Light yellow = minimum relative humidity for the given year, Dark green = maximum relative humidity for the given year

	Mean daily temperature															
	17-Apr	**18-Apr**	**19-Apr**	**20-Apr**	**21-Apr**	**22-Apr**	**23-Apr**	**24-Apr**	**25-Apr**	**26-Apr**	**27-Apr**	**28-Apr**	**29-Apr**	**30-Apr**		
2014	12.25	18.15	15.35	13.6	13.7	13.7	17.75	19.7	12.65	14.5	19	16.95	16.7	21.1		
2015										19.14	22.5	13.8	15.65	16.15		
2016						20.49	16.12	5.82	7.39	12.85	**5**	11.1	15.2	18.2		
2017									18.4	16.9	11.7	**7.4**	8.3	12.8		
2018													23.12	22.55		
2019					20.97	15.61	9.42	18	23.9	**25.9**	13.3	11.4	12.2	15.1		
	01-May	**02-May**	**03-May**	**04-May**	**05-May**	**06-May**	**07-May**	**08-May**	**09-May**	**10-May**	**11-May**	**12-May**	**13-May**	**14-May**	**15-May**	
2014	20.7	20.7	16.2	12.8	12.8	16.1	18.9	18.5	19.7	19.1	14	14.7	13.3	11	**9.8**	
2015	16.15	13.9	**12.1**	17.8	23.05	25.05	21.95	21.3	21.65	19.15	19.7	22.4	22.7	19.15	13.2	
2016	17.1	17.7	15.2	17.05	17.5	20.8	19.6	17.95	18.8	11.25	15.35	15.4	15	14.7	10.85	
2017	14.2	15	14.1	13.1	13.8	15.3	13.7	15.2	9.1	12.8	14.4	16.2	15.9	17.5	20.5	
2018	22.85	23.85	24.55	**24.9**	24.95	22.45	21.7	21.15	19.65	22.6	22.2	23.75	22.45	20.25	**13.55**	
2019	18.5	19.2	14.6	10.2	9	**5.5**	12	15.4	8.9	15.5	19	17.2	18.1	6.6	10.3	
	16-May	**17-May**	**18-May**	**19-May**	**20-May**	**21-May**	**22-May**	**23-May**	**24-May**	**25-May**	**26-May**	**27-May**	**28-May**	**29-May**	**30-May**	**31-May**
2014	11.4	14.1	13.4	20.25	21.35	23.5	**25.35**									
2015	21.3	18.5	21.1	26.35	**26.7**	12.3	12.9	15.45	19.7	15.55	17.35	14	14.3	21.7	23.35	
2016	12.9	10.6	14.6	17.4	19.9	22.95	24.25	**24.85**	14.9	22.65	23.3	24	23.05	24.9	21.85	23.65
2017	21.8	21.2	22.05	**23.6**	20.75	21.2	23.5	21	17.9	16.8	20.5	21.85	22.45	23.55		
2018	16.4	16.75	18.95	19.9	20.55	21.55	20.68	22.78	22.37							
2019	10.4	15.5	19.6	19.6	16.9	17.9	12.9	14	18.3	22	22.6	17.9	17	17.2	12.8	18.6
	01-Jun	**02-Jun**	**03-Jun**	**04-Jun**												
2014																
2015																
2016	21.7	16.44	18.78													
2017																
2018																
2019	22.9	23.06	25.6	22.76												

Mean daily realive humidity																
	17-Apr	**18-Apr**	**19-Apr**	**20-Apr**	**21-Apr**	**22-Apr**	**23-Apr**	**24-Apr**	**25-Apr**	**26-Apr**	**27-Apr**	**28-Apr**	**29-Apr**	**30-Apr**		
2014	64.3	51.1	64.05	71.3	89.4	95.5	79.55	67.75	96.6	**97.65**	63.85	75.05	72.7	51.45		
2015										57	44.75	89.7	44.1	46.6		
2016						42.62	53.42	78.89	61.96	**40.7**	90.65	58.45	49	43.6		
2017									52	63.7	85.2	**93**	80	73.9		
2018													70	69.9		
2019					**24.59**	54.67	84.58	69.3	44.1	39.4	67.2	72	74.9	68.3		
	01-May	**02-May**	**03-May**	**04-May**	**05-May**	**06-May**	**07-May**	**08-May**	**09-May**	**10-May**	**11-May**	**12-May**	**13-May**	**14-May**	**15-May**	
2014	48.15	58	75.15	78.4	61.25	59.8	61.2	54.25	59.15	**45.25**	95.3	59.6	71.9	83.95	92.9	
2015	62.7	82.9	86.1	90.05	71.7	63.95	44.85	45.55	52.9	50.3	45.05	**40.95**	55.9	59	77.05	
2016	67.1	70.8	73.25	65.45	63.4	50.15	51.85	57.9	58.15	**94**	87.3	89.55	92.55	89.85	64.1	
2017	62.2	84.5	83.9	88	85.8	83.8	77.1	64.5	68.4	59.6	68.4	82.5	70.5	78.1	60.3	
2018	50.15	64.05	54.95	57.65	57.65	52.25	**40.1**	54.15	79.95	59.05	65.1	57.15	44.95	48.25	**95.05**	
2019	42.9	48.9	66.5	92.1	**99.3**	98.1	51.6	50.9	97.5	70.7	58.5	77.8	67.2	94.9	98.1	
	16-May	**17-May**	**18-May**	**19-May**	**20-May**	**21-May**	**22-May**	**23-May**	**24-May**	**25-May**	**26-May**	**27-May**	**28-May**	**29-May**	**30-May**	**31-May**
2014	95.35	82.35	87.1	63.45	61.9	59.65	57.45									
2015	50.7	54.45	53.55	47	52.2	96.9	**97.6**	97.4	72.8	88.6	83.9	60	60.2	49.25	52.55	
2016	57.3	70.75	61.55	67.8	54	47.05	51.75	54.2	90.9	61.5	60.45	64.65	72.05	67.05	73.3	56.95
2017	56.8	57.75	59.05	53.6	64.05	65.8	58.7	70.65	78.15	69.25	52.65	52.55	**47**	50.52		
2018	72.3	72.75	70.1	64.35	59	51.9	74.77	68.09	70.32							
2019	90.6	75.4	65.2	72.6	81.9	67.5	94.4	81.7	70.8	50.7	60.4	83.7	92.5	89.2	97.3	76.7
	01-Jun	**02-Jun**	**03-Jun**	**04-Jun**												
2014																
2015																
2016	61.15	89.9	88.86													
2017																
2018																
2019	58.8	67.34	57.79	68.6												

Appendix 3: Overview of the most supported models and the significant covariates of the Cormack-Jolly-Seber model selection.

These models were selected based on their AICc values and likelihood ratio tests. Blue or red indicates (-) negative or (+) positive effects of variables on apparent survival (Phi) or recapture probability (p). DOF: Day of the flight period.

		Phi		p	
		The most supported models	**Variables with significant effects**	**The most supported models**	**Variables with significant effects**
2014	males	~Age (-) + temperature (-)	DOF (-) time thorax (+)	~time	DOF (-) temperature (+) relative humidity (-) mass (+) thorax (-)
	females	~DOF (-) + relative humidity (+)	Age (-) thorax (+)	~relative humidity (-) + thorax (+) + thorax^2 (-)	time temperature (+)
2015	males	~DOF (-) + thorax (+)	Age (-) DOF (-) thorax^2 (-)	~observers (+) + Cohort (-) + wing (+) + wing^2 (-)	time temperature (+) relative humidity (-) thorax (-)
	females	~DOF (-)	Age (-) temperature (+) relative humidity (-) thorax (+)	~observers (+) + temperature (+) + thorax (+)	time relative humidity (-) wing (+)
2016	males	~DOF (-) + temperature (-)	Age (-) Cohort (-) time relative humidity (+) proboscis (+)	~time	DOF (-) observers (+) temperature (+) relative humidity (-)
	females	~Age (-) + temperature (-)	DOF (-) time	~Age (+) + relative humidity (-)	time observers (+) temperature (+) mass (+) thorax (+)

		Phi		p	
		The most supported models	Variables with significant effects	The most supported models	Variables with significant effects
2017	males	~DOF (-) + relative humidity (+)	Age (-) time temperature (-) wing (+) proboscis (+)	~time	DOF (-) Cohort (-) observers (+) temperature (-) relative humidity (-)
	females	~DOF (-) + relative humidity (+)	Age (-) time temperature (-)	~time	relative humidity (+)
2018	males	~DOF (-)	Cohort (-) temperature (-) thorax (+)	~Cohort (-) + relative humidity (-)	DOF (-) temperature (+) mass (-) thorax (+) thorax^2 (+) wing (+)
	females	~DOF (-)	temperature (-) relative humidity (+) thorax (+)	~time	temperature (+) relative humidity (-) thorax (+)
2019	males	~Age (-) + temperature (-) + wing (+)	DOF (-) time relative humidity (+) mass (+) mass^2 (-) thorax (+) wing (+) wing^2 (-) proboscis (+)	~time	observers (+) temperature (+) relative humidity (-) mass (+) mass^2 (-) thorax (+)
	females	~Age (-) + temperature (-) + mass (+)	DOF (-) time relative humidity (+) thorax (+) thorax^2 (-)	~temperature (+)	DOF (+) Time Cohort (+) observers (+) relative humidity (-) mass (-) wing (-) proboscis (-)

Appendix 4: AICc and estimates of Beta coefficients (with 95% CI) of the six most supported models’ covariates of the Cormack-Jolly-Seber model selection.

These models were selected based on their AIC values and likelihood ratio tests. DOF: Day of the flight period, observers: Number of observers, factor variable with 3 levels, thorax: thorax width, wing: wing length, proboscis: proboscis length. In ‘p(time)’ models a separate coefficient (Beta) was estimated for each sampling occasion.

Year	**Sex**	**Model**	**Model AICc**	**Phi**		**p**	
				Covariate	**Beta**	**Covariate**	**Beta**
2014	**male**	**Phi(Age+temperature) p(time)**	**1449.412**	**Age**	**-0.148 (-0.192 — -0.103)**	**time**	
				temperature	**-0.117 (-0.210 — -0.024)**		
		Phi(Age) p(time)	1453.388	Age	-0.125 (-0.167 — -0.083)	time	
		Phi(DOF + temperature) p(time)	1469.218	DOF	-0.109 (-0.143 — -0.076)	time	
				temperature	-0.126 (-0.200 — -0.052)		
		Phi(DOF) p(time)		DOF	-0.070 (-0.110 — -0.029)	time	
		Phi(Age + temperature + thorax) p(temperature+thorax)	1477.405	Age	-0.167 (-0.207 — -0.126)	temperature	0.213 (0.155 — 0.273)
				temperature	-0.146 (-0.221 — -0.071)	thorax	-1.138 (-2.143 — -0.134)
				thorax	1.460 (0.141 — 2.779)		

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2014	**male**	Phi(Age + temperature + thorax) p(temperature + wing + wing2)	1477.883	Age	-0.167 (-0.207 — -0.126)	temperature	0.209 (0.150 — 0.268)
				temperature	-0.147 (-0.222 — -0.072)	wing	-7.160 (-8.278 — -6.042)
				thorax	1.406 (0.088 — 2.724)	wing^2	0.115 (0.096 — 0.133)
	female	**Phi(DOF + relative humidity)** **p(relative humidity + thorax + thorax2)**	**927.591**	**DOF**	**-0.125 (-0.177 — -0.072)**	**relative humidity**	**-0.051 (-0.069 — -0.033)**
				relative humidity	**0.027 (-0.003 — -0.072)**	**thorax**	**34.940 (14.292 — 55.588)**
						thorax^2	**-8.496 (-13.725 — -3.266)**
		Phi(DOF) p(relative humidity + thorax + thorax2)	929.018	DOF	-0.109 (-0.161 — -0.056)	relative humidity	-0.050 (-0.068 — -0.032)
						thorax	35.732 (15.564 — 55.900)
						thorax^2	-8.702 (-13.816 — -3.589)
		Phi(DOF + relative humidity) p(time)	929.199	DOF	-0.131 (-0.180 — -0.083)	time	
				relative humidity	-0.029 (0.0007 — 0.058)		
		Phi(Age) p(relative humidity + thorax + thorax2)	930.305	Age	-0.111 (-0.165 — -0.057)	relative humidity	-0.049 (-0.067 — -0.031)
						thorax	35.880 (15.945 — 55.815)
						thorax^2	-8.708 (-13.766 — -3.651)
		Phi(DOF) p(time)	931.584	DOF	-0.070 (-0.110 — -0.029)	time	
		Phi(Age) p(time)	934.251	Age	-0.097727 (-0.154 — -0.042)	time	

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2015	**male**	**Phi(DOF + thorax) p(observers + Cohort + wing + wing^2)**	**907.721**	**DOF**	**-0.147 (-0.206 — -0.089)**	**observers: level 2**	**1.183 (0.527 — 1.839)**
				thorax	**3.113 (1.043 — 5.183)**	**observers: level 3**	**2.141 (1.312 — 2.971)**
						Cohort	**-0.076 (-0.144 — -0.008)**
						wing	**15.934 (15.487 — 16.380)**
						wing^2	**-0.262 (-0.270 — -0.254)**
		Phi(DOF + thorax) p(observers + Cohort + proboscis + proboscis^2)	909.815	DOF	-0.152 (-0.211 — -0.093)	observers: level 2	1.172 (0.517 — 1.826)
				thorax	2.883 (0.822 — 4.943)	observers: level 3	2.116 (1.296 — 2.936)
						Cohort	-0.094 (-0.164 — -0.024)
						proboscis	19.566 (17.404 — 21.728)
						proboscis^2	-0.813 (-0.904 — -0.722)
		Phi(DOF + thorax) p(observers + wing + wing^2)	910.470	DOF	-0.150 (-0.208 — -0.092)	observers: level 2	1.235 (0.581 — 1.888)
				thorax	3.116 (1.057 — 5.176)	observers: level 3	2.190 (1.362 — 3.019)
						wing	17.090 (16.669 — 17.511)
						wing^2	-0.281 (-0.289 — -0.273)
		Phi(Age) p(observers + Cohort + wing + wing^2)	911.491	Age	-0.149 (-0.210 — -0.088)	observers: level 2	1.175 (0.520 — 1.829)
						observers: level 3	2.121 (1.296 — 2.947)
						Cohort	-0.093 (-0.162 — -0.024)
						wing	17.121 (16.716 — 17.526)
						wing^2	-0.281 (-0.288 — -0.274)
		Phi(Age) p(observers + Cohort + proboscis + proboscis^2)	912.653	Age	-0.155 (-0.217 — -0.094)	observers: level 2	1.165 (0.512 — 1.818)
						observers: level 3	2.090 (1.276 — 2.904)
						Cohort	-0.113 (-0.184 — -0.041)
						proboscis	20.682 (18.592 — 22.771)
						proboscis^2	-0.858 (-0.946 — -0.770)
		Phi(DOF + thorax) p(time)	912.974	DOF	-0.154 (-0.212 — -0.096)	time	
				thorax	3.011 (0.995 — 5.027)		

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2015	**female**	**Phi(DOF)** **p(observers + temperature + thorax)**	**815.497**	**DOF**	**-0.120 (-0.177 — -0.063)**	**observers: level 2**	**1.166 (0.509 — 1.823)**
						observers: level 3	**2.068 (1.262 — 2.873)**
						temperature	**0.142 (0.079 — 0.205)**
						thorax	**1.839 (0.609 — 3.070)**
		Phi(DOF) p(observers + temperature + wing)	817.469	DOF	-0.120 (-0.177 — -0.063)	observers: level 2	1.176 (0.514 — 1.838)
						observers: level 3	2.119 (1.307 — 2.932)
						temperature	0.149 (0.084 — 0.213)
						wing	0.244 (0.054 — 0.434)
		Phi(DOF + thorax) p(observers + temperature)	818.477	DOF	-0.112 (-0.169 — -0.055)	observers: level 2	1.150 (0.494 — 1.805)
				thorax	1.655 (0.262 — 3.048)	observers: level 3	2.071 (1.264 — 2.879)
						temperature	0.145 (0.081 — 0.209)
		Phi(relative humidity) p(observers + temperature + thorax)	819.394	relative humidity	-0.033 (-0.051 — -0.016)	observers: level 2	1.247 (0.603 — 1.892)
						observers: level 3	2.297 (1.488 — 3.105)
						temperature	0.126 (0.065 — 0.189)
						thorax	2.051 (0.880 — 3.222)
		Phi(relative humidity + thorax) p(observers + temperature + wing)	821.971	relative humidity	-0.027 (-0.044 — -0.009)	observers: level 2	1.262 (0.613 — 1.911)
				thorax	1.551 (0.088 — 3.014)	observers: level 3	2.330 (1.515 — 3.144)
						temperature	0.135 (0.072 — 0.199)
						wing	0.220 (0.035 — 0.406)
		Phi(DOF + thorax) p(time)	827.001	DOF	-0.096 (-0.175 — -0.016)	time	
				thorax	1.809 (0.245 — 3.373)		

Year	**Sex**	**Model**	**Model AICc**	**Phi**		**p**	
				Covariate	**Beta**	**Covariate**	**Beta**
2016	**male**	**Phi(DOF + temperature)** **p(time)**	**1433.756**	DOF	-0.125 (-0.167 — -0.082)	time	
				temperature	-0.105 (-0.172 — -0.040)		
		Phi(DOF + relative humidity) p(time)	1436.587	DOF	-0.158 (-0.197 — -0.120)	time	
				relative humidity	0.029 (0.006 — 0.051)		
		Phi(DOF) p(time)	1441.378	DOF	-0.169 (-0.207 — -0.132)	time	
		Phi(Age + temperature) p(time)	1445.177	Age	-0.096 (-0.131 — -0.061)	time	
				temperature	-0.174 (-0.236 — -0.111)		
		Phi(Age + relative humidity + proboscis) p(time)	1464.648	Age	-0.125 (-0.159 — -0.090)	time	
				relative humidity	0.035 (0.012 — 0.058)		
				proboscis	0.486 (0.052 — 0.920)		
		Phi(DOF + temperature) p(DOF + temperature)	1470.321	DOF	-0.124 (-0.166 — -0.082)	DOF	-0.041 (-0.066 — -0.015)
				temperature	-0.110 (-0.175 — -0.045)	temperature	0.157 (0.109 — 0.205)
	female	**Phi(Age + temperature)** **p(Age + relative humidity)**	**1082.251**	**Age**	**-0.1 (-0.151 — -0.050)**	**Age**	**0.050 (0.008 — 0.091)**
				temperature	**-0.105 (-0.174 — -0.037)**	**relative humidity**	**-0.040 (-0.054 — -0.026)**
		Phi(Age + temperature) p(time)	1090.557	Age	-0.094 (-0.144 — -0.044)	time	
				temperature	-0.106 (-0.175 — -0.036)		
		Phi(Age) p(relative humidity)	1093.992	Age	-0.126 (-0.173 — -0.078)	relative humidity	-0.041 (-0.055 — -0.027)

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2016	female	Phi(Age + temperature) p(observers + temperature)	1094.252	Age	-0.096 (-0.146 — -0.047)	observers: level 2	1.008 (0.363 — 1.653)
				temperature	-0.104 (-0.173 — -0.036)	observers: level 3	1.084 (0.305 — 1.862)
						temperature	0.114 (0.069 — 0.159)
		Phi(temperature) p(Age + relative humidity)	1095.854	temperature	-0.151 (-0.218 — -0.085)	Age	0.043 (0.001 — 0.084)
						relative humidity	-0.040 (-0.054 — -0.026)
		Phi(Age) p(Age + observers + temperature)	1096.266	Age	-0.132 (-0.179 — -0.084)	Age	0.063 (0.017 — 0.108)
						observers: level 2	1.146 (0.481 — 1.811)
						observers: level 3	1.383 (0.566 — 2.201)
						temperature	0.104 (0.059 — 0.150)
2017	male	**Phi(DOF + relative humidity) p(time)**	**1207.887**	**DOF**	**-0.153 (-0.217 — -0.09)**	**time**	
				relative humidity	**0.045 (0.001 — 0.089)**		
		Phi(DOF) p(time)	1210.027	DOF	-0.188 (-0.239 — -0.138)	time	
		Phi(Age + relative humidity) p(time)	1211.295	Age	-0.091 (-0.138 — -0.044)	time	
				relative humidity	0.092 (0.050 — 0.134)		
		Phi(Age + temperature) p(time)	1219.440	Age	-0.089 (-0.135 — -0.042)	time	
				temperature	-0.178 (-0.268 — -0.088)		
		Phi(relative humidity) p(time)	1222.285	relative humidity	0.117 (0.074 — 0.160)	time	

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2017	**male**	Phi(Age + relative humidity + wing) p(DOF + observers + relative humidity)	1296.173	Age	-0.105 (-0.151 — -0.058)	DOF	-0.136 (-0.180 — -0.092)
				relative humidity	0.085 (0.043 — 0.128)	observers: level 2	0.786 (0.317 — 1.255)
				wing		observers: level 3	1.774 (1.083 — 2.465)
						relative humidity	-0.071 (-0.095 — -0.047)
	female	**Phi(DOF + relative humidity) p(time)**	**1002.359**	**DOF**	**-0.168 (-0.233 — -0.103)**	**time**	
				relative humidity	**0.053 (0.002 — 0.104)**		
		Phi(DOF) p(time)	1004.616	DOF	-0.186 (-0.247 — -0.125)	time	
		Phi(Age + temperature) p(time)	1009.985	Age	-0.068 (-0.125 — -0.012)	time	
				temperature	-0.249 (-0.384 — -0.114)		
		Phi(temperature) p(time)	1013.329	temperature	-0.284 (-0.413 — -0.154)	time	
		Phi(DOF + relative humidity) p(relative humidity + proboscis + proboscis2)	1014.673	DOF	-0.161 (-0.226 — -0.096)	relative humidity	-0.018 (-0.037 — 0.001)
				relative humidity	0.052 (0.002 — 0.101)	proboscis	-12.861 (-15.087 — -10.636)
						proboscis^2	0.536 (0.444 — 0.629)
		Phi(Age + relative humidity) p(time)	1015.101	Age	-0.086 (-0.141 — -0.031)	time	
				relative humidity	0.097 (0.033 — 0.161)		

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2018	male	**Phi(DOF p(Cohort + relative humidity)**	**1391.050**	**DOF**	**-0.068 (-0.116 — -0.020)**	**Cohort**	**-0.221 (-0.298 — -0.144)**
						relative humidity	**-0.048 (-0.062 — -0.033)**
		Phi(DOF) p(time)	1396.407	DOF	-0.071 (-0.120 — -0.021)	time	
		Phi(~1) p(Cohort + relative humidity)	1396.583	~1		Cohort	-0.232 (-0.308 — -0.156)
						relative humidity	-0.050 (-0.064 — -0.034)
		Phi(~1) p(Cohort + temperature)	1397.027	~1		Cohort	-0.187 (-0.265 — -0.109)
						temperature	0.257 (0.179 — 0.334)
		Phi(thorax) p(time)	1398.129	thorax	1.499 (0.267 — 2.732)	time	
		Phi(Cohort) p(time)	1399.315	Cohort	-0.080 (-0.151 — -0.008)	time	
	female	**Phi(DOF) p(time)**	**1132.199**	**DOF**	**-0.097 (-0.164 — -0.030)**	**time**	
		Phi(~1) p(time)	1135.290	~1		time	
		Phi(time) p(relative humidity)	1143.099	time		relative humidity	-0.033 (-0.048 — -0.018)
		Phi(time) p(time)	1143.890	time		time	
		Phi(time) p(temperature)	1152.075	time		temperature	0.118 (0.046 — 0.190)
		Phi(DOF) p(relative humidity)	1154.974	DOF	-0.080 (-0.136 — -0.024)	relative humidity	-0.033 (-0.048 — -0.018)

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2019	**male**	**Phi(Age + temperature + wing)** **p(time)**	**1394.179**	**Age**	**-0.111 (-0.144 — -0.078)**	**time**	
				temperature	**-0.076 (-0.130 — -0.021)**		
				wing	**0.324 (0.149 — 0.500)**		
		Phi(Age + wing) p(time)	1399.388	Age	-0.113 (-0.146 — -0.08)	time	
				wing	0.325 (0.151 — 0.499)		
		Phi(Age + temperature + body mass + body mass^2) p(time)	1400.685	Age	-0.108 (-0.141 — -0.076)	time	
				temperature	-0.074 (-0.128 — -0.019)		
				body mass	54.234 (9.022 — 99.446)		
				body mass^2	-117.076 (-226.253 — -7.899)		
		Phi(Age + temperature + proboscis) p(time)	1400.975	Age	-0.104 (-0.136 — -0.072)	time	
				temperature	-0.076 (-0.130 — -0.022)		
				proboscis	0.481 (0.105 — 0.858)		
		Phi(Age + temperature + thorax) p(time)	1401.420	Age	-0.101 (-0.133 — -0.070)	time	
				temperature	-0.073 (-0.126 — -0.019)		
				thorax	2.061 (0.372 — 3.749)		
		Phi(Age + temperature + wing) p(observers + temperature + body mass + body mass^2)	1407.753	Age	-0.113 (-0.146 — -0.080)	observers: level 2	1.373 (0.749 — 1.998)
				temperature	-0.087 (-0.140 — -0.035)	observers: level 3	2.015 (1.289 — 2.740)
				wing	0.312 (0.136 — 0.488)	temperature	0.161 (0.098— 0.224)
						body mass	69.534 (28.408 — 110.661)
						body mass^2	-149.414 (-248.615 — -50.212)

Year	Sex	Model	Model AICc	Phi		p	
				Covariate	Beta	Covariate	Beta
2019	**female**	**Phi(Age + temperature + body mass) p(temperature)**	**950.915**	**Age**	**-0.138 (-0.185 — -0.091)**	**temperature**	**0.285 (0.212 — 0.358)**
				temperature	**-0.209 (-0.292 — -0.125)**		
				body mass	**8.719 (2.574 — 14.864)**		
		Phi(Age + temperature + body mass) p(Cohort + temperature)	952.952	Age	-0.138 (-0.185 — -0.091)	Cohort	-0.002 (-0.028 — 0.024)
				temperature	-0.209 (-0.292 — -0.125)	temperature	0.286 (0.211 — 0.362)
				body mass	8.712 (2.567 — 14.857)		
		Phi(Age + temperature + thorax) p(temperature)	954.946	Age	-0.128 (-0.173 — -0.083)	temperature	0.285 (0.212 — 0.358)
				temperature	-0.215 (-0.297 — -0.133)		
				thorax	2.027 (0.022 — 4.031)		
		Phi(Age + temperature) p(temperature)	956.913	Age	-0.118 (-0.161 — -0.075)	temperature	0.284 (0.211 — 0.357)
				temperature	-0.222 (-0.304 — -0.140)		
		Phi(DOF + temperature) p(temperature)	964.751	DOF	-0.077 (-0.111 — -0.042)	temperature	0.286 (0.213 — 0.359)
				temperature	-0.145 (-0.227 — -0.063)		
		Phi(Age + temperature + body mass) p(time)	974.159	Age	-0.140 (-0.188 — -0.093)	time	
				temperature	-0.231 (-0.326 — -0.136)		
				body mass	8.375 (2.155 — 14.595)		

Appendix 5: Results of GOF tests.

Yellow markings indicate ĉ -hat adjustment for those data. (DOF=day of the flight period, RH=relative humidity, Temp=temperature, tw=thorax width, wing=wing length, obs=level of observers)

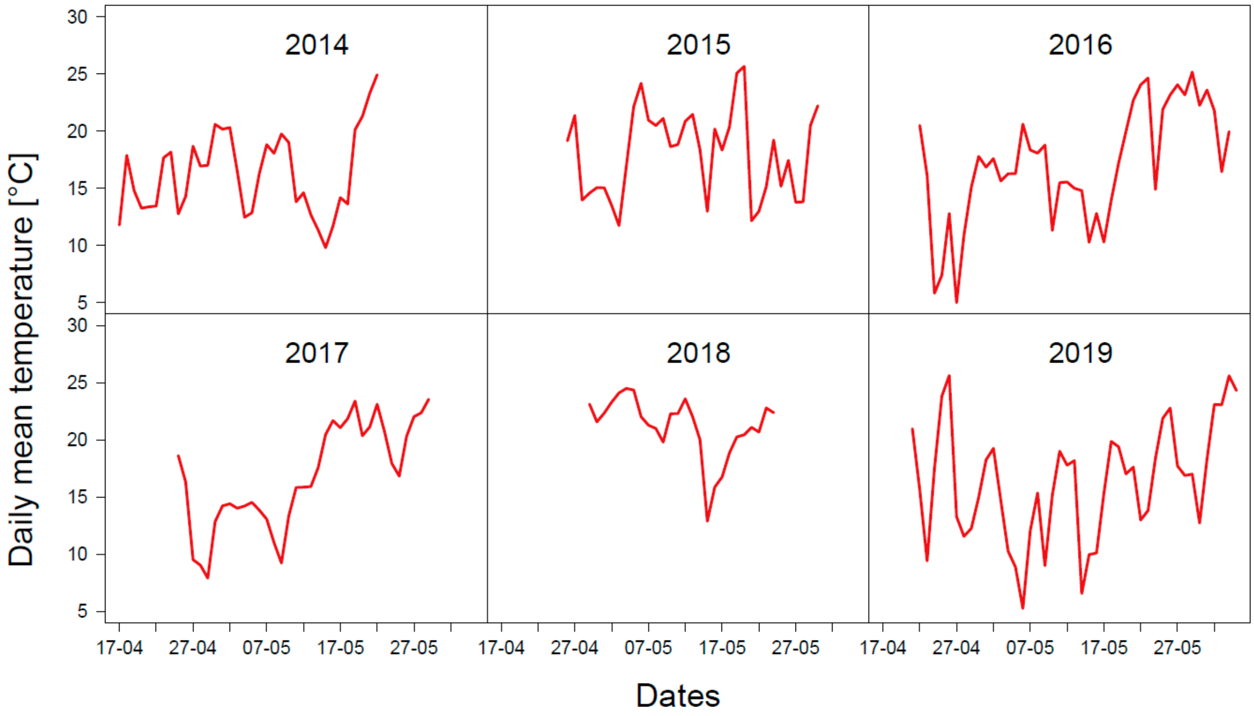
Model	Used method	Method of estimating ĉ	2014		2015		2016		2017		2018		2019	
			females	males	females	males	females	males	females	males	females	males	females	males
Phi(time) p(time)	MARK: Release GOF	**χ2 / d.f.**	1.074	0.714	0.957	1.033	0.972	0.939	0.774	**1.637**	**1.385**	**1.512**	0.716	**1.590**
	p-value of Release GOF		0.337	0.942	0.555	0.415	0.537	0.603	0.872	0.003	0.033	0.019	0.951	0.004
	MARK: Bootstrap GOF	**Model Deviance / Mean Deviance**	1.013	0.970	0.967	1.026	0.983	1.003	1.082	1.055	1.078	1.059	0.966	**0.866**
		model ĉ / mean ĉ	1.746	0.989	1.981	1.137	1.092	1.238	2.454	**1.709**	**1.222**	1.039	1.038	0.910
	U-CARE	**χ2 / d.f.**	1.134	0.647	0.824	1.054	0.854	0.834	0.684	**1.370**	**1.476**	**1.498**	0.786	**1.461**
		Global Test (significant p value)								**YES**	**YES**	**YES**		**YES**
Phi(Age) p(time)	MARK: Bootstrap GOF	**Model Deviance / Mean Deviance**	1.018	0.945	1.005	1.009	0.980	1.010	1.088	1.071	**1.103**	1.043	0.944	**0.865**
		model ĉ / mean ĉ	0.879	0.967	1.292	1.094	1.029	1.069	1.477	**1.268**	**1.235**	0.982	0.995	0.933
Phi(DOF) p(time)	MARK: Bootstrap GOF	**Model Deviance / Mean Deviance**	0.985	0.935	0.951	0.961	0.944	0.968	1.045	1.035	1.089	1.043	0.880	**0.809**
		model ĉ / mean ĉ	1.069	0.940	1.225	1.025	0.957	1.014	1.301	**1.182**	**1.201**	1.017	0.874	**0.862**
Most supported models (PHI)			DOF +RH	Age +temp	DOF	DOF +tw	Age +temp	DOF +temp	DOF +RH	DOF +RH	DOF	DOF	Age +temp +mass	Age +temp +wing
Significant variables (PHI)			Age, DOF	Age, DOF, time	Age, DOF	Age, DOF	Age, DOF, time	Age, DOF, time	Age, DOF, time	Age, DOF, time	DOF	DOF	Age, DOF, time	Age, DOF, time
Most supported models (P)			RH +tw+tw^2	time	obs +temp+tw	obs +Cohort +wing+$wing^2$	Age +RH	time	time	time	time	Cohort +RH	temp	time
Significant variables (P)			time	DOF, time	time	time	Age, time	DOF, time	time	DOF, time	time	DOF	DOF, time	time

Appendix 6: χ^2, degree of freedom and p-value of TEST 2 and 3 of the RELEASE GOF TESTS in 2017 and 2019 for males and in 2018 for both sexes.

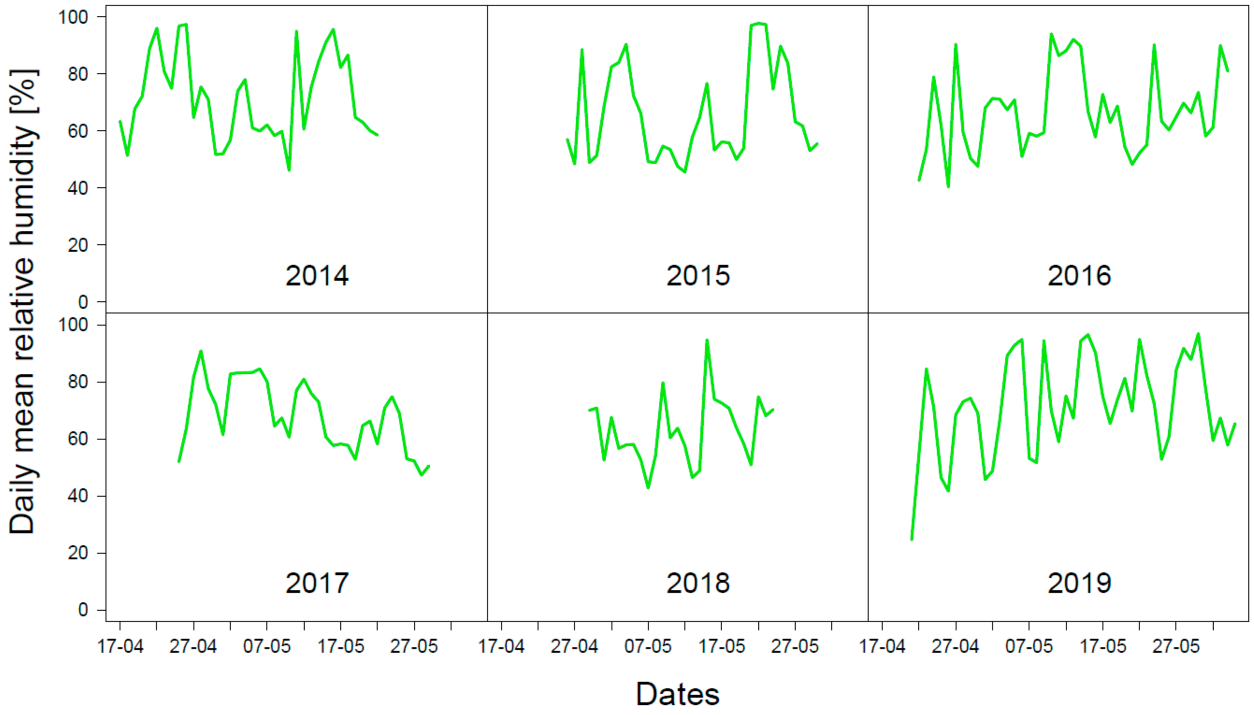
	Release GOF Tests	χ^2	d.f	p-value
2017 MALES	TEST 3.Sm6	6.02	1	0.0141
	TEST 3.Sm14	7.87	1	0.0050
	TEST 3.Sm15	6.54	1	0.0105
	TEST 3.Sm30	4.00	1	0.0455
	TEST 2.C7	4.17	1	0.0412
	TEST 2.C10	5.14	1	0.0233
	TEST 2.C11	20.58	1	0.0000
	TEST 2.C12	4.84	1	0.0279
	TEST 2.C14	8.33	1	0.0039
	TEST 2.C21	7.92	1	0.0049
	TEST 2.C22	7.74	2	0.0209
	TEST 2.C25	5.61	1	0.0178
	TEST 2.C27	7.00	1	0.0082
2018 FEMALES	TEST 3.Sm2	4.00	1	0.0455
	TEST 3.Sm7	8.36	1	0.0038
	TEST 3.Sm13	5.83	1	0.0157
	TEST 2.C3	3.94	1	0.0472
	TEST 2.C4	5.96	1	0.0146
	TEST 2.C10	9.05	3	0.0286
	TEST 2.C12	6.56	2	0.0376
2018 MALES	TEST 3.SR4	6.48	1	0.0109
	TEST 3.SR8	5.28	1	0.0215
	TEST 3.Sm5	8.02	1	0.0046
	TEST 3.Sm8	5.27	1	0.0217
	TEST 3.Sm10	4.24	1	0.0396
	TEST 3.Sm11	27.00	1	0.0000
	TEST 2.C11	6.20	1	0.0127
	TEST 2.C19	3.94	1	0.0472
2019 MALES	TEST 3.SR8	9.37	1	0.0022
	TEST 3.SR12	15.48	1	0.0001
	TEST 3.Sm7	4.62	1	0.0316
	TEST 3.Sm11	5.51	1	0.0190
	TEST 2.C4	11.98	1	0.0005
	TEST 2.C8	5.05	1	0.0247
	TEST 2.C11	6.98	2	0.0305
	TEST 2.C13	4.00	1	0.0455
	TEST 2.C14	10.98	1	0.0009
	TEST 2.C18	5.17	1	0.0230
	TEST 2.C19	9.85	1	0.0017
	TEST 2.C20	7.48	1	0.0063
	TEST 2.C26	4.00	1	0.0455

Appendix 7: Mean daily temperature (a) and relative humidity (b) during flight periods for all years.

a)



b)



Acknowledgements

I would like to thank my supervisor, Dr. János Kis, for providing the opportunity and professional support for this topic.

I express my gratitude to my supervisor, Dr. Ádám Kőrösi, for his endless help and patience in the planning and analysis of my research. I am grateful for his continuous support throughout my work with his professional advice and ideas.

I thank both of them, as well as Ádám Gór, Dr. Viktor Szigeti, Dr. Flóra Vajna, and the students of the University of Veterinary Medicine for their help in the planning, preparation, review of joint publications, and field sampling. I am also grateful to Dr. Flóra Vajna for providing us with her research funding, which supported the material and equipment needs of our research. I would like to thank the Duna-Ipoly National Park Directorate for their assistance. I also appreciate the feedback from the reviewers and editors of my articles.

I am grateful to my opponents, Prof. Dr. Tibor Szép and Dr. Zsolt Végvári, for the time and effort they invested. Thanks for their high-quality reviews, their insightful critical comments, and useful suggestions.

I would like to thank my parents, grandparents, sister, and brother-in-law for encouraging me and providing unconditional support throughout my studies.

Finally, I am deeply grateful to my partner, Ádám Gór, for his endless patience, support, and professional advice.