# Investigating the current occupancy of the endangered butterfly *Parnassius apollo* and the abundance of its host plant *Sedum telephium* in the Archipelago Sea

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Insect populations across the globe are declining and the biodiversity of insects is threatened. To deal with the threats, conservation biology aims to identify species that are most vulnerable to extinction, and also to understand mechanisms that make some species more prone than others to population decline, range contraction and extinction. For the extinction risk assessments it is crucial to study and monitor the possible changes in the numbers and ranges of species. Furthermore, it is important to examine the drivers of declines. This knowledge sheds light to the nature of extinction processes and enables conservation management planning, as the most threatened species often require specific management and conservation measures. Out of all insect groups declining worldwide, butterflies (Lepidoptera) are among the taxa most affected. In Finland butterflies are considered as one of the most endangered organism groups. Among the endangered butterfly species is the Apollo butterfly (*Parnassius apollo*), which has declined in numbers throughout the species range since the 1970s. Two decades ago the Apollo and its sole host plant orpine (*Sedum telephium*) was studied in one of its few strongholds in southwest Finland in the Archipelago Sea. The host plant was found to be the most important factor affecting the occupancy of the Apollo larvae. Today the species and the study area are included in CoastNet LIFE project that plans to implement restoration work of habitat for the Apollo butterfly. For successful restoration work, it is important to update the data on the Apollo and its host plant. In this thesis I study the possible occupancy changes of the Apollo larvae and possible changes in their host plant abundance by comparing historical data from survey years 1997, 1999-2003 to the data I collected in the same study area in 2019. I also explore if abundance of *S. telephium* affects the occupancy of *P. apollo* larvae. To examine the probable change in the larvae occurrence I compared naïve occupancy estimations and occupancy model estimates between data sets of historical years and the year 2019. In the historical data the number of *S. telephium* plants per site were scored in categories: 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). I used these same categories to study the possible change in the abundance of *S. telephium* in the comparison between earlier survey years and the year 2019. I found a very strong decline in the occupancy of the larvae and no apparent difference in the abundance of their host plant between the historical data and the data I collected 2019. However, there were difference between survey sites. The Apollo larvae were mainly detected on sites with average amount of the host plant (10-100 plants) instead of sites with high abundance of the host plant. This finding is unexpected as it suggests that the abundance of the host plant is not as important as was predicted based on previous studies. In addition, I did tentative grouping of survey sites with k-means cluster analysis. I included the island groups (0-3) to top occupancy model and the analyze showed a clear difference between the groups. This difference between island groups indicates the importance of the spatial location to the probability of occupancy of the Apollo larvae. In light of these results, it is critical to continue monitoring the endangered Apollo larvae in order to understand if the decline is merely a temporary change or a signal of a possible trend. Additionally, for successful conservation management it is important to research the drivers affecting the population decline.

KEYWORDS: Parnassius, Apollo, butterfly, endangered, larvae, host plant, occupancy, telephium, endangered, archipelago

# Contents

# 1 Introduction

## 1.1 Biodiversity and butterflies

There are many different measures of biodiversity. Essentially, biodiversity reflects the number, variety, and variability of living organisms. The degree of biodiversity loss is commonly measured with the use of species extinction rates. Today's rapid rate of extinctions indicates that our planet is facing a catastrophic loss of biodiversity (Ceballos et al., 2020). Restoring and protecting biodiversity is important because Earth's ecosystems and human welfare rely on it. Therefore, nearly all countries have a legislation that obligates them to classify and protect threatened species (Mattila, 2008). Furthermore, one of the 20 Aichi Biodiversity Targets set by Convention on Biological Diversity was "By 2020 the extinction of known threatened species has been prevented and their conservation status, particularly of those most in decline, has been improved and sustained" (CBD, 2012). To fulfill these obligations and targets, conservation biology is seeking to identify species and populations that are most vulnerable to extinction (Bergen et al., 2019). However, it is a challenging task. In addition, conservation biology is attempting to discover and understand processes that make some species more susceptible than others to population decline, range contraction and extinction (Caughley, 1994; Pimm et al., 1988). This knowledge is used to shed light to the nature of extinction processes and to plan conservation management (Mattila, 2008), as most threatened species often require specific management and conservation measures (Rassi et al., 2010).

For successful conservation management scientist must collect data, e.g., abundance and distribution on species, as management and conservation decisions that could save a species or population is often limited due to lack of adequate survey data to monitor wildlife (Evangelista et al., 2012). However, in population biology resources are generally limited for collection of data and thus, field research is often done in short periods and in restricted areas (Ehrlich & Murphy, 1987; O'Grady et al., 2004). Limited resources and short-term surveys present a problem, since conservation biology is by definition seeking solutions to preserve populations and species in the long term (Ehrlich & Murphy, 1987). Nevertheless, as Ehrlich and Murphy (1987) presented already in their time that the brief histories of single populations over short periods repeatedly have proven to give at best a partial, and at worst an incorrect, impression of the factors controlling the distribution and abundance of different species. Nowadays, due to the urgency with many threatened species wildlife managers are exploring new and more efficient methods to collect data and to assess a population's range and distribution, recognize critical habitats, and

instruct conservation priorities (Evangelista et al., 2012). Several of the new models and techniques are disciplined by presence and absence data in association with environmental data to statistically characterize a species' ecological niche within a landscape (Evangelista et al., 2012). Today data can be collected, e.g., with large-scale sensors or satellite remote sensing and aerial imagery as well as land-based observation platforms that can increase efficiency, standardization, and the value of monitoring data for various uses (Chandler et al., 2017). However, human observations offer important calibration and validation for the remotely collected data and assessments (e.g., species distribution models) (Evangelista et al., 2012). In case of herbivorous insects, Ehrlich and Murphy (1987) concluded already in their time that the size of reserves required to preserve these species is often underestimated. They suggested that conservation biologists would gain important information and benefit from long-term monitoring of populations of sample invertebrate groups. Indeed, today conservation biology has benefited from long-term studies of ecological model systems that have recognized factors affecting population declines and extinctions (Bergen et al., 2019). Nonetheless, there are still inadequate data available for most threatened species (O'Grady et al., 2004).

Parmesan (2003) presented that butterflies (Lepidoptera) are ideal model system for understanding effects of modern environmental changes. For the reason that, unlike in any other studied taxa, in butterflies have changes across the whole species ranges been documented (Parmesan, 2003). Parmesan (2003) added that as a model system, butterflies are key organisms for monitoring of biodiversity. Moreover, butterflies are among the scarce wild organisms that can provide quality information of climate change. This is due to the sensitivity of butterflies to climate, the temporal and spatial extent of distributional data, and the abundance of biological knowledge available, especially of European species (Chandler et al., 2017; Parmesan, 2003). For instance, distributions of several Lepidoptera species living in the boreal zone have shifted towards the north, and the effects of climate change are evident in the data, especially for species living in fells (Cerrato et al., 2019; Hyvärinen et al., 2019). Additionally to views of Parmesan, Ehrlich (2003) described that butterflies have become a valuable model system for increasing basic knowledge e.g., in ecology, evolutionary and conservation biology. Ehrlich (2003) agreed that butterflies meet the criteria for well-known indicator taxa and added that butterflies can give us clues about the conservation value of various areas and help monitoring the health of ecosystems chosen for preservation (Ehrlich, 2003). Besides, butterflies can serve as umbrella species - species that are used for making conservation-related decisions (Ehrlich, 2003). In addition, conservation of umbrella species is likely to result in the conservation of other organisms as well via protecting

certain areas (Ehrlich, 2003). Thus, protecting butterflies in particular areas could also benefit several other species.

## 1.2   Conservation criteria

The ongoing destructive loss of biodiversity that Earth is experiencing demands us to identify species that are at risk quickly and efficiently (O'Grady et al., 2004). Nonetheless, lack of direct estimates of extinction risk and adequate data for most species present a problem (O'Grady et al., 2004). One solution to the problem is to use alternative indicators in  the classifications of threatened species (O'Grady et al., 2004). Alternative indicators include biological parameters, such as, population size, temporal variability of population size, range size, fragmentation of range, body size and life history specialization which are generally associated with extinction proneness (O'Grady et al., 2004). In addition to various parameters used, there are several protocols invented to estimate and categorize the degree of species extinction risk and conservation status (Mattila, 2008). One of these systematic protocols and universally used for ranking taxa is the International Union for Conservation of Nature (IUCN) red list. The IUCN Red List Categories and Criteria were developed to estimate species extinction risk in the international level (IUCN, 2003; IUCN, 2019). IUCN extinction risk evaluations are based on quantitative criteria that categorize species based on their relative extinction risk (IUCN, 2019). Criteria require data on e.g., population size, rates of decline and fragmentation of habitat (IUCN, 2019). For instance, the IUCN Red List of Threatened Species describe that most vulnerable species to climate change are species with specialized habitat or microclimate requirements, narrow environmental tolerances, dependence on environmental cues or interspecific interactions, and poor dispersal ability (IUCN, 2008).

For butterflies in Finland the IUCN Red List classification is predominantly based on the area of occupancy or distribution of species (Rassi et al., 2001). For instance, criterion A used in the Red List evaluation is reduction in population size (Rassi et al., 2010). The criterion A state that "reduction in population size must be observed, estimated, inferred, or suspected over the last ten years or three generations (whichever is the longer), where the causes of reduction are clearly reversible and understood and ceased based on, e.g., direct observation and/or a decline in area of occupancy, extent of occurrence and/or quality of habitat" (Rassi et al., 2010). Other criteria include, e.g., criterion C; small and continuously declining population which comprehends fewer than 2500 mature individuals for 'endangered' and either continuing estimated at least 20 % decline within five years or two generations or continuing decline in number of mature individuals with additional conditions, see Rassi et al., 2010 for more details, and criterion D; very small or restricted population which comprehends population size estimated to number fewer

than 250 mature individuals for 'endangered' (Rassi et al., 2010). In consideration of the criteria for IUCN classification, it is crucial to study the numbers and range of species, and monitor the possible changes. Besides the IUCN classification, population trends provide valuable information for conservation biology (O'Grady et al., 2004). This information can be used to determine population viability and conservation status (O'Grady et al, 2004). Additionally, population trends makes it possible to specify conservation priorities and plan management even before species begin to decline (Mattila, 2008).

## 1.3 Trends in butterflies

Van Swaay & Warren (1999) already presented in their Red Data Book of European Butterflies, following the IUCN criteria, that during 20th century butterflies have suffered significant declines in population abundance, range contractions and numerous extinctions around Europe. The issue is very relevant also today, as a recent review by Sánchez-Bayo & Wyckhuys (2019) exhibited striking rates of insect declines that could lead to the extinction of 40 % of the world's insect species over the next few decades. They discovered that the main drivers of insect species decline appeared to be, e.g., habitat loss and conversion to intensive agriculture, pollution, biological factors, and climate change. Sánchez-Bayo & Wyckhuys (2019) presented that in terrestrial ecosystems Lepidoptera (with Hymenoptera and Coleoptera) appear to be the taxa most affected. Furthermore, the affected groups not only include specialist, but also many common and generalist species (Sánchez-Bayo & Wyckhuys, 2019). As insects comprise nearly two thirds of all terrestrial species on Earth, the declining trends are profoundly affecting life forms everywhere (Sánchez-Bayo & Wyckhuys, 2019). Furthermore, the substantial insect declines could trigger wide-ranging cascading effects within many of the world's ecosystems (Sánchez-Bayo & Wyckhuys, 2019). Hence, the issue is serious and concerns the whole planet.

A similar trend is observed in Finland where nearly half of the native butterfly species have suffered from the changes in the environment since the 1970s, and in 1976 the first butterflies protected by law were the Apollo butterfly (*Parnassius apollo,* Linnaeus 1758) and clouded apollo (*Parnassius mnemosyne*, Linnaeus 1758) (Marttila et al., 1991). Moreover, according to the most recent Red List assessment of Finnish species, 30.3 % (n = 716) of the assessed Lepidoptera taxa was red listed (Hyvärinen et al., 2019). In addition, the group of Lepidoptera was placed among the organism groups with quantitatively highest number of threatened species (Hyvärinen et al., 2019). Thus, butterflies are considered as one of the most endangered organism groups also in Finland.

Butterflies are vulnerable group due to their specific habitat requirements (Crone & Schultz, 2003; van Swaay & Warren, 1999). Most butterflies inhabit restricted and special ecological niches (Crone & Schultz, 2003; van Swaay & Warren, 1999). For these species the world can be divided into habitat and non-habitat (Crone & Schultz, 2003; van Swaay & Warren, 1999). In addition to specific habitat requirements, many butterflies are restricted not only to just one or two foodplants and to types of vegetation, but also to successional stages (van Swaay & Warren, 1999). More than twenty years ago, Van Swaay & Warren (1999) reported that almost 90 % of threatened European butterfly species are affected, among various other threats, by widespread loss and reduction in size of breeding habitats. The threat is ongoing, as according to the 2019 Red List of Finnish species the most important habitats of threatened and near threatened butterfly species are sun-exposed environments, forest edges or in semi-open forests and shore habitats, which are continuing to decline (Hyvärinen et al., 2019). Moreover, the most serious threats to Lepidoptera species are the disappearance of habitats through overgrowing of open areas or through various land-use changes (Hyvärinen et al., 2019). The loss and reduction in size of suitable breeding habitats is causing habitat isolation and fragmentation (van Swaay & Warren, 1999). Hence, many butterflies are considered to exist and behave like metapopulations which rely on networks of habitat patches (van Swaay & Warren, 1999; Crone & Schultz, 2003).

Metapopulations are described as a set of subpopulations with a dynamic balance between colonization and extinction of the patches; where extinction events depend on population size, and colonization is outlined by an immigration from other patches (Hanski et al., 1995). However, the balance may be disturbed for many butterfly species due to habitat loss (Crone & Schultz, 2003). Furthermore, due to habitat fragmentation when a local population goes extinct, the patch is often too isolated to be recolonized (Thomas, 1991). Thus, butterflies that leave a habitat patch are likely to be lost, rather than finding a new patch suitable for reproducing (Crone & Schultz, 2003). Subsequently, many butterfly populations today must survive independently of other patches, if they are to persist at all (Crone & Schultz, 2003). Reduced population sizes are likely to decrease genetic diversity, which may make extinction more probable due to inbreeding (Saccheri et al., 1998). In addition, the difficulty of finding suitable new sites to colonize makes even small changes in climate more likely to have greater effects than in the past (Parmesan, 2003). Before single populations may have been larger and lived-in better-connected habitat network and/or in bigger, unbroken habitat patches and thus, populations could shift location (Parmesan, 2003). Dallas et al. (2020) added that besides habitat patch area and connectivity, metapopulation studies should address the influence of local environmental conditions and/or the spatial arrangement of habitat

patches to the metapopulation dynamics. Therefore, extensive information on species occurrence and habitat requirements is critical when predicting species distributions and planning conservation strategies (Dallas et al., 2020; Eilers et al., 2013).

Mattila (2008) investigated ecological traits that predispose Finnish butterflies to extinction risk and found that larval specificity promotes the risk of extinction. In more detail, species specified to sole host plant type are dependent on local abundance of host plants (Warren, 1992) and their inability to switch host plants makes them more sensitive to changes. Furthermore, host plant distribution in monophagous species predicts extinction risk (Kotiaho et al., 2005). Mattila (2008) assumed that it may be because species cannot be more widely distributed than their host plants, and because extinction risk assessments of butterflies are generally based on distribution (Rassi et al., 2001).

## 1.4   Occupancy estimation and modeling to study wildlife

Prediction of species' distributions and species distribution models are essential to various applications in ecology, evolution, and conservation science (Tyre et al., 2003). For instance, knowledge of species' distributions is crucial for instructing conservation action and for effective conservation planning (Guisan & Thuiller, 2005). Therefore, observing and counting individuals to estimate their abundance and distribution has a long tradition in ecology and management (MacKenzie et al., 2018d). For instance, species richness and occupancy (proportion of area occupied a species or fraction of landscapes units where the species is present) are parameters that could be used to evaluate the status of community or population (MacKenzie et al., 2018d). However, examining population abundance trends is often difficult for most species (Ewing & Gangloff, 2016). There is generally limited availability of historical information on population sizes, and resources are often inadequate to generate contemporary abundance estimates (Ewing & Gangloff, 2016; Pollock, 2006). This information shortage frequently leads to decisions about conservation status based on informal data (Ewing & Gangloff, 2016). In many cases the only available information of species previous occurrences is presence records from databases where people share observations or from museum collections (Guillera-Arroita, 2017). It could be speculated that the purposes of such data are probably to document the distribution or presence of a species or groups of species instead of determining abundance (Ewing & Gangloff, 2016). Furthermore, usually the data available are prone to estimation biases due to lack of information about sampling effort (Guillera-Arroita, 201). From such data it is not possible to know if the scarce species records are because of species rarity or because of little sampling effort (Guillera-Arroita, 2017). Thus, data sets that include information

about survey effort are much less susceptible to estimation biases and can examine species occurrence probabilities (Guillera-Arroita, 2017). Such data sets include not only species presence but also absence records (Guillera-Arroita, 2017). However, when considering species distribution modeling presence-absence data are a robust method and do not account for detectability (Guillera-Arroita, 2017; Lahoz-Monfort et al., 2014). Detectability is an important variable, as two most common and important types of errors emerging in ecological surveys are false negatives and false positives (Chen et al., 2013; Guillera-Arroita, 2017). Especially false negatives are the most common error in species occurrence data and arise when species are not detected in surveys of occupied sites (Guillera-Arroita, 2017). Species detectability can vary in space and/or time and even sessile species are often detected imperfectly (Chen et al., 2013; Guillera-Arroita, 2017). Thus, the problem with false absences is worth the attention (Guillera-Arroita, 2017). If observation errors in the data are ignored, the induced estimates of species distributions can be biased, even when species detectability is constant (e.g., Guillera-Arroita, 2017; Lahoz-Monfort et al., 2014; Tyre et al., 2003). Thus, Lahoz-Monfort et al. (2014) recommended that consideration of imperfect detection should be consistently taken into account in species distribution modelling.

Methodological toolbox for estimation of species distribution fortunately have evolved over the years and today there are methods that permit conclusions about occupancy based on detection non-detection data and account for imperfect detection (MacKenzie et al., 2018d). However, like all data used for species distribution modelling, detection non-detection data should include information about the detection process with the information if species was detected or not in a set of surveyed sites (Guillera-Arroita, 2017). As mentioned earlier, species detectability can vary but there are also various factors affecting species detectability (Guillera-Arroita, 2017). These include, e.g., species and habitat characteristics, abundance, surveyor skills or detection method, survey effort and survey conditions (Guillera-Arroita, 2017). Hence, one way to improve the precision of the estimated distribution is to reduce the problem of imperfect detection (Guillera-Arroita, 2017). The common error of false absences can be corrected, e.g., by increasing the amount visits to the sites (repeated surveys) or by using a more sensitive detection method (Guillera-Arroita, 2017). This extra survey effort and information is used in the models to better estimate the species' distribution from process noise and observation error (Guillera-Arroita, 2017). These models were mostly developed to be used in wildlife monitoring and are known as 'occupancy models' (Guillera-Arroita, 2017). Occupancy model by definition is a model that takes imperfect detection of organisms in surveys into account, and is used to estimate the probability of true presence or absence of a organism at a site (Stanford University). The occupancy model is created by

determining the detection probability of a species at a site based on collected data (Stanford University).

Occupancy modeling offers a flexible framework to explore ecological questions and processes (Bailey et al., 2014;, Guillera-Arroita, 2017). Therefore, it is beneficial for ecologists who study and analyze, e.g., species distribution models, range dynamics, habitat relationships, abundance data and metapopulation dynamics (Bailey et al., 2014; Guillera-Arroita, 2017). For example, Boyce et al. (2016) presented a theory that predicts the occupancy of a species will decrease as the habitat quality decrease. In this framework, it is possible to describe the sensitivity of a species as a change in the occupancy along the range of ecosystem attributes if stressors affecting species occupancy are identified (Cortelezzi et al., 2017).

## 1.5   The Apollo butterfly and its host plant

PhD Marianne Fred together with PhD Jon Brommer have done pioneering research of the Apollo butterfly (*Parnassius apollo)* and its habitat patches with possible stressors affecting the species occupancy during years 1997 and 1999-2003 (Brommer & Fred, 1999; M. S. Fred et al., 2006; M. S. Fred & Brommer, 2003, 2005, 2009, 2015). A great part of the research was done in one of few Apollo's strongholds in southwest Finland within the Archipelago national park in the Archipelago Sea. Fred & Brommer (2009) studied the structure of the *P. apollo* population based on dispersal in the Archipelago study area and noticed that nearly 80 % of the adult butterflies stay on their natal island. In addition, the average turnover rate base on larval occupancy was 8 % per year (Fred & Brommer, 2009). In the light of these results they concluded that the population can be viewed as a metapopulation of island populations (Fred & Brommer, 2009).

Globally *P. apollo* is classified as vulnerable (Dixon, 1996). However, the assessment needs updating (Dixon, 1996). In Finland *P. apollo* is currently classified as an endangered (EN) species (Hyvärinen et al., 2019). According to Fred and Brommer (2015) it is not clear why the Apollo have become locally extinct in many parts of Finland. However, Fred and Brommer (2005, 2003) suggested that *P. apollo* is a habitat specialist that depends on small scale heterogeneity in the distribution of adult and larval resources. Thus, loss of habitat has most likely been a considerable threat to it (Fred & Brommer, 2005, 2003). The Apollo butterfly may become extinct unless the circumstances that are threatening its survival and reproduction improve (Rassi et al. 2010).

Orpine (*Sedum telephium*) is the sole host plant for the *P. apollo* larvae in Finland and islands where it grows provide a possible habitat patch for the Apollo. However, *S. telephium* is a common species in rocky outcrops in South Finland and therefore it is not

a limiting a factor for *P. apollo* larvae occurrence. Nevertheless, Fred and Brommer (2003) found that the amount and the density of host plant on a patch is the most important habitat parameter and critical factor for *P. apollo* larvae and their survival in the archipelago. Thus, monitoring the host plant patches is crucial for *P. apollo* population, since conservation of the butterflies depends on securing host plant *S. telephium* abundance on large enough patches. Additionally, Fred and Brommer (2003) also found that patch occupancy is more reliable in describing the *P. apollo* population if density of larvae found per patch is low.

## 1.6  Aims of this study

CoastNet LIFE project was launched in 2018 aiming to improve the conservation status of Natura 2000 sites along the Baltic coastal zone. The project is managed by Metsähallitus nature services and it plans to implement restoration work on different types of habitat, e.g., boreal Baltic islets and small open habitats during period of 2018-2025. The restoration work is expected to help increase the populations of numerous species by giving them new areas to colonize and to help battle the effects of habitat fragmentation and isolation. Among CoastNet LIFE's various expected results is restoration of habitat (50 ha) in the Archipelago national park area for the Apollo butterfly whose population is expected to grow threefold. The project's knowledge on the Apollo butterfly is mostly based on studies made nearly 20 years ago by Fred and Brommer in the Archipelago national park study area. This study aims to add contemporary occupancy data to the previous knowledge from survey years 1997, 1999-2003, referred as historic data.

The aims of this study are 1) to collect information on the occupancy of *P. apollo* larvae and compare it to historic occupancy, 2) to study the current abundance of *S. telephium* and evaluate whether a change has occurred during the last two decades, 3) to explore if abundance of *S. telephium* affects the occupancy of *P. apollo* larvae.

My hypotheses for this study are first, that the occupancy of *P. apollo* larvae has changed, possibly due to climate change, as the observed trend of Lepidoptera species living in the boreal zone shifting towards the north (Cerrato et al., 2019; Hyvärinen et al., 2019). Second, my hypothesis is that abundance of *S. telephium* has declined because it grows on biotopes that are vulnerable to overgrowth (Hyvärinen et al., 2019; Kontula & Raunio, 2018). Third and finally, I presume that the *P. apollo* larvae will not occupy survey sites where *S. telephium* is not abundant, because based on earlier work the abundance of the host plant is considered to be the most important factor affecting the occupancy of the larvae (Fred & Brommer, 2003), and as Boyce et al. (2016) presented that the occupancy of a species will decrease as the habitat quality decrease.

To my knowledge, this is the first time that the *P. apollo* occupancy is examined at this scale after Fred and Brommer's research. This study will shed light on the contemporary status of the previously known stronghold of *P. apollo* in the Archipelago Sea. This study highlights on possible occupancy changes in *P. apollo* larvae that is crucial information for the CoastNet LIFE project to plan the management and to focus the effort of restoring habitats on appropriate sites.

## 2  Material and methods

### 2.1  Study species

#### 2.1.1  *Parnassius apollo*

*Parnassius apollo* (Linnaeus, 1758) is a member of the Papilionoidea family, inhabiting montanous areas in the palearctic region (Marttila et al., 1991). Although the Apollo occurs in wide geographical scope from west Europe to central Asia, it generally occurs only in low densities (Marttila et al., 1991). In Finland the species distribution is restricted to the archipelago and the south-west coast (Marttila et al., 1991).

 As mentioned before, Fred & Brommer (2009) studied the structure of the *P. apollo* population based on dispersal in islands of the Archipelago Sea study area. The islands form a network of differently sized habitat patches in the archipelago (Fred, 2004). Fred and Brommer (2009) suggested that the population can be considered as island populations forming a metapopulation.

The Apollo butterfly is large butterfly (wingspan around 65-95 mm) and a good flyer, even in windy conditions (Collins & Morris, 1985; Marttila et al., 1991). It has rounded, milky white wings with black spots, grey markings and the hindwings have outstanding scarlet spots (Haahtela et al., 2011). The species is univoltine, the larvae and adult butterflies occur between late April and September, however, in mountains and in the northern range *P. apollo* occurs from May to August (Marttila et al., 1991). The Apollo overwinters in an egg-state and larvae hatch in the spring and start feeding on their host plant during warm and sunny days (Fred & Brommer, 2003). Apollo larvae occur only once in the vegetation and pupate after three to four weeks (Fred & Brommer, 2003). Due to the high mobility of the adult Apollo butterflies, the presence of larvae is the only reliable sign of host-plant patch having been used for breeding (Fred et al., 2006). The larvae development is not synchronized and during the larval period, there are some individuals that still have not hatched from the egg and those that have already pupated (Fred & Brommer, 2003). Female Apollo butterflies do not oviposit in the immediate surroundings of the host plant, as many other butterfly species, thus making the egg and pupal stages difficult to detect (Fred & Brommer, 2003). However, larvae that are 0.5 cm or longer can

be detected due to their conspicuous colorization and because they leave signs of grazing (Fred & Brommer, 2003). The early instars are velvet black, but as the larvae grow the protuberances on the sides of each segment develop a distinct red color that act as warning color and a defense mechanism (Fig. 1) (Tolman & Lewington, 1997). As palearctic species, the larvae can endure sudden drops in the temperature by producing chemical substances that prevent freezing (Marttila et al., 1991). Nevertheless, the black coloration may be critical for heat absorbance, since the larvae sun-basking behavior is common (Bohlin et al., 2008; Marttila et al., 1991). The larvae hide in the undergrowth when they are not feeding and can therefore be difficult to detect, but the signs of grazing can be used as an indicator of locations worth re-visiting if the larvae are not seen at once (Fred & Brommer, 2003). However, there are several other organisms also forage on *S. telephium* and may leave signs of grazing.



**Figure 1.** Two pictures of a *Parnassius apollo* larva grazing on a *Sedum telephium* stem, the larva's sole host plant in Finland. In the picture on the left the leaves of the stem are damaged by the grazing. The larvae can reach a length of 6 cm before pupating.

Globally the IUCN Red List classification for *P. apollo* is vulnerable, however, this evaluation was done in the 20th century and thus, may be outdated (Dixon, 1996). In Finland, the Apollo was most abundant in the 1930s but started declining markedly in 1940s and has been protected by law since 1976 (Marttila et al., 1991). Today the Apollo population in Finland is considered to be stable but rare (Finnish Biodiversity Info Facility). However, the species have become locally extinct in many parts of Finland and the Red List classification has changed from nearly threatened to endangered in the last two evaluations (Hyvärinen et al., 2019; Rassi et al., 2001; Rassi et al., 2010). According to Fred and Brommer (2015) it is not clear why the Apollo is declining. However, they suggested that loss of habitat has most likely been a considerable threat to it (Fred & Brommer 2005, 2003). As mentioned before, the circumstances that are threatening

survival and reproduction of *P. apollo* has to improve or the species may become extinct (Rassi et al. 2010). Therefore, monitoring the host plant patches is crucial for *P. apollo* population, since conservation of the butterflies depends on securing host plant abundance on large enough patches.

## 2.1.2 *Sedum telephium*

*Sedum telephium* (Linné, 1753)*,* belonging to the family of Crassulaceae, is a perennial herb native to areas of temperate climatic conditions and a common species in rocky outcrops in South Finland (Fig. 2) (Blamey et al., 2005). The rocky outcrops are usually partly forested and represent extremely poor, dry habitat (Brommer & Fred, 1999; Kontula & Raunio, 2018). The species also known as orpine or livelong is easily detected in spring as it is among the earliest plants to have shoots after winter (Fred & Brommer, 2009). In the Archipelago Sea in south-western Finland *S. telephium* grows typically in small groups on barren rocky islands and shores. These nutrient-poor biotopes are extremely vulnerable to nitrogen deposition (Weiss, 1999) and suffer from eutrophication of the Baltic Sea and overgrowth (Hyvärinen et al., 2019; Kontula & Raunio, 2018; Rassi et al., 2010). Thus, *S. telephium* could be used as an early warning signal of overgrowth in rocky islands endangered biotopes (Kontula & Raunio, 2018). Furthermore, *S. telephium* is the sole host plant for the *P. apollo* larvae in Finland. Thus, islands where *S. telephium* grows provide a possible habitat patch for the Apollo larvae. According to Fred and Brommer (2010, 2003, 1999) the amount and the density of host plant on a patch is the most important habitat parameter and critical factor for *P. apollo* larvae and their survival in the archipelago.
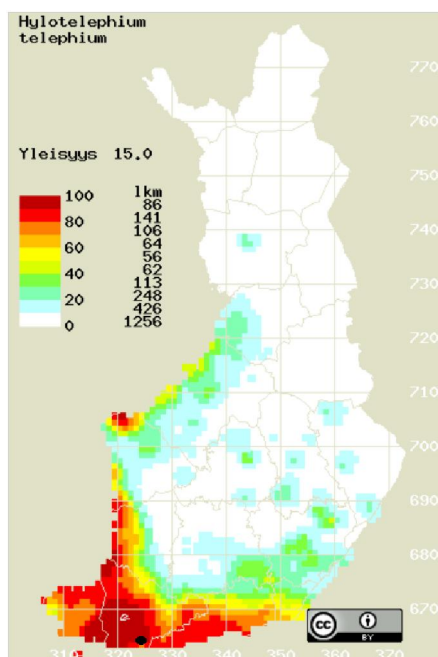


**Figure 2.** Frequency of *Sedum (or Hylotelephium) telephium* in Finland (Lampinen & Lahti, 2019). The darker color indicates higher frequency. The species is common in the southwest of Finland. The black dot represents the approximate location of the study area in the Archipelago Sea.

## 2.2 Study area

This study was conducted in southwest Finland within the Archipelago national park in the Archipelago Sea. This archipelago is one of the world's largest and it is located between the Åland island and mainland Finland (Metsähallitus, 2018). According to Metsähallitus (2018) the national park itself includes more than 2,000 islands and rugged, rocky skerries, and the more extensive co-operation area has approximately 8,400 islands and islets separated by open waters. Moreover, 93 % of the national park area consists of water. The entire study area, including open water areas was approximately 165 km$^2$ (Fig. 3).
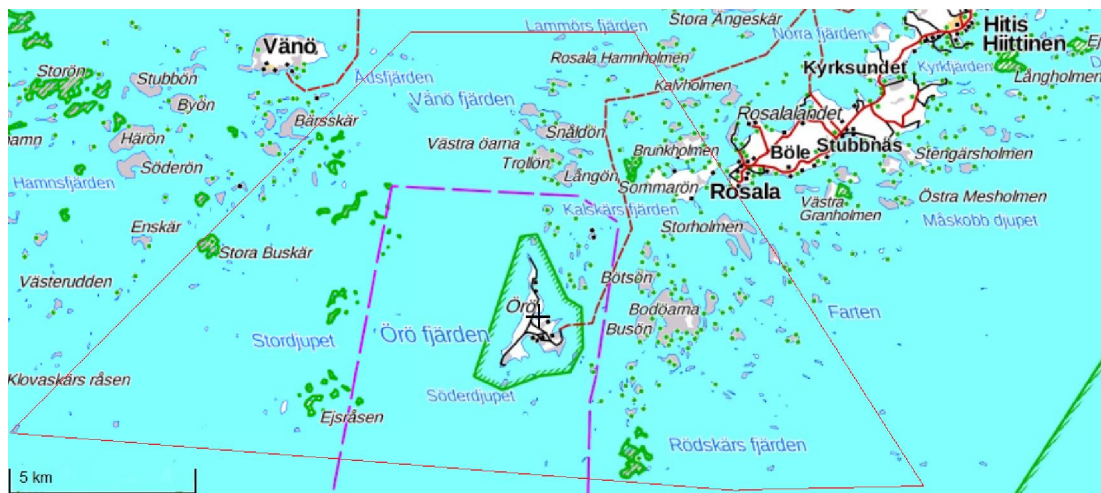


**Figure 3.** The study area in the Archpelago Sea. The approximate study area is bordered with thin solid red lines. The green color indicates areas of the Archipelago national park (National Land Survey of Finland, MapSite).

PhD Marianne Fred together with PhD Jon Brommer have studied the *P. apollo* habitat patches during years 1997 and 1999-2003 in the same area, referred as the historical data (Brommer & Fred 2007, 1999, 1998; Fred & Brommer 2015, 2009, 2005, 2003; Fred et al. 2006; Fred 1997). In the year 1997 more than 170 islands were surveyed, and variable subset of islands were monitored 1999-2003. For this study, a subset of 85 islands were selected from a larger set of islands surveyed in 1997. The selection was based on the historical occupancy of *P. apollo* and abundance of *S. telephium*. The subset of 85 islands was considered as the main survey sites for this study. However, after surveying the main 85 islands due to favorable weather conditions and time remaining, I was able to collect data from additional 33 islands in the Archipelago national park and its co-operation area. I collected data total from 118 different islands. Most of the surveyed islands were the same as in the year 1997.

The surveyed islands vary in size and vegetation. Some of the larger islands are partly forested and/or junipers (*Juniperus communis*) are dominating the island, and the

smallest very barren islets have sparse patches of vegetation. The barren islands vegetation is mainly composed of grasses and shrubs with surrounding areas of mosses, lichens, and bare rock. The surveyed area within the islands was focused in the zone between junipers and the sea where the host plant *S. telephium* grows (referred as edge band) and thus, the area surveyed was considered suitable for *P. apollo* larvae.

## 2.3 Data collection

The historical data from previous survey years (1997 and 1999-2003) included the coordinates of 78 survey islands and a copy of a map with documented previous survey locations in the Archipelago national park study area. The data consisted of yearly information of survey islands; 1997 (n = 174), 1999 (n = 85), 2000 (n=83), 2001 (n=84), 2002 (n=83) and 2003 (n = 74). The information also included the island's *S. telephium* class category; 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants), the number of detected *P. apollo* larvae and occupancy status of the site, the survey date and time spent per site and weather condition during the survey (sunny, cloudy and/or windy). This data acted as a baseline for my study.

I had two main objectives when collecting data for this thesis. First, to estimate the current occupancy of *P. apollo* larvae in the survey islands and second, to estimate the current abundance of *S. telephium* plants. I collected the data in order to compare the results to the historical data and to study possible changes in the occupancy and abundance of the survey species, the larvae and the host plant. Together with four team members I collected data periodically from 16th of May until 8th of June 2019 in the Archipelago national park study area. I was present every time we went and collected data. Other members of the team participated in different days or weeks, when it was possible for them. Usually there were at least two people collecting the data and generally we were able to visit 8-10 survey sites per day. To maximize the comparability of the collected data to the historical data I was instructed the survey method used in previous surveys by Fred and Brommer (2005, 2003). However, Fred and Brommer (2003) did not conduct repeated surveys as in this study. They tested the survey accuracy of their studies by conducting removal counts. Fred and Brommer (2003) counted and temporarily removed all observed larvae from a set of patches during three subsequent surveys. These removal counts addressed whether a patch scored as empty, truly was empty (Fred & Brommer, 2003). In addition, the removal counts allowed Fred and Brommer (2003) to make direct comparisons on larval abundance between two populations they were studying, the coastal population and the archipelago population. Furthermore, the removal counts served as validation of their previous data on larval abundance based on single visit. Fred and Brommer (2003) found that the survey method was reliable, especially in the archipelago population where the density of larvae

was high. However, they also found that if the density of larvae detected per patch is low as in the coastal population, patch occupancy is more reliable in describing the population.

In the beginning of each survey, name of the island, date, starting time and weather conditions (sunny, windy, foggy and/or cloudy), how many people and who were surveying, number of *S. telephium* plants and detected *P. apollo* larvae were recorded. In addition, ending time of the survey, if other species of Lepidoptera larvae were detected and if damaged plants that possibly were grazed by any larvae were observed were recorded.

Most of the study islands were surveyed by two people, one walking clockwise and the other counterclockwise around the shoreline of the island until the surveyors met. The surveys focused on the zone between junipers and the sea where *S. telephium* grows (the edge band). There were also checks done to the inner part of the island on some occasions. Inner parts were checked often when there were three observers, and one could walk the center line of the island while two observers were going around. Walking different routes, towards each other's reduced the probability of double counting. Additionally, it decreased possible disturbance of incubating birds to minimal. However, there were few occasions when two people were walking in the same direction. This was done when the survey island's edge band was very wide, and it was considered more efficient to cover to whole edge band width at the same time by two people walking apart from each other's counting own part of the edge band. In these situations, to prevent double counting the surveyed area was divided using available landmarks. In addition, there were two occasions when a new person joined the data collecting team and walked with another, more experienced person. New team member rehearsed the method by counting the same plant individuals as the more experienced person. Thus, two people were walking in the same direction and one against. In these occasions, the result of people walking in the same direction were documented and mean value calculated for analyses. Furthermore, 47 times out of 165 surveys were done by only one person. Large part of solo surveys were due to limited survey time and a few because of the small size of the island. Solo surveys saved time because often there was no need to anchor the boat and the boat driver could drop off team members to different islands.

The host plants on the entire island were counted. However, as mentioned before, the counting concentrated most times on the edge band. The checks done to inner part of larger sites revealed that the method of focusing the counting on edge band was efficient. Counting of *S. telephium* was done by counting whole plants instead of individual stems. However, small separately growing stems were also calculated, and a group of five stems was documented as one plant individual. Each observer's individual count and the

total count of the island was documented. In the historical data the number of *S. telephium* plants per site were scored in categories: 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). These same categories were used in the data collected in 2019. This was done in order to compare the score to the historical score. In addition, the categories were used to give a *S. telephium* class value for each survey island to make comparison between sites. A patch was considered suitable for *P. apollo* larvae if host plants occurred. All suitable patches were surveyed with approximately equal efforts per unit area, when searching for *S. telephium*.

People surveying *S. telephium* plants additionally counted the number of detected *P. apollo* larvae and measured them. The signs that larvae leave when grazing are useful aid for detecting, since damaged plants are generally conspicuous even from a distance and suggest the presence of a larva nearby. However, there are many other species that also consume *S. telephium,* and drought and/or diseases may damage the plants. Thus, no excess time were used for searching *P. apollo* larvae if a thorough visual examination of the surroundings of a damaged plant did not result in detection of a larva. The total amount of time surveying *P. apollo* larvae on each island depended on the total amount of host plant present.

After every survey day I gathered the data from every person that had participated in the surveys and I assigned each surveyed island *S. telephium* abundance score (0-4) and *P. apollo* larvae occupancy status not detected = 0 or detected = 1.

Directly after I had the data from the firstly surveyed main survey sites of 85 islands, I chose 55 from the main islands for a second (repeated) survey. These repeated surveys were planned in order to estimate the accuracy of the method and to estimate the detection probability and to correct possible observation errors for occupancy modeling (MacKenzie et al., 2018e). Since, generally the problem with scoring presence/absence is the alleged empty patches, especially with a species where the larvae occur singly such as *P. apollo* (Fred & Brommer, 2003). The second surveys were conducted on the way to new, additional survey sites. I chose the 55 islands for repeated survey from all four compass points to cover most of the study area and where we could reach within the limited time frame. In addition to location, I selected the 55 sites based on their history of occupancy, current occupancy status, weather conditions, number of observed damaged plants on the first survey in 2019 as well as on their location in the study area. I included islands where we had and had not detected larvae on the first survey, and which had been considered occupied at least one year in the historical data. If the first survey had been done in sub-optimal weather conditions (cloudy), the repeated survey was done in optimal weather conditions (sunny) to increase the detection probability of the larvae. We were able to conduct a repeated survey on 44 (or nearby) of the planned

55 islands. To increase the independency of the repeated surveys they were done in a different day, different time of the day and by different observer, as the first survey occasion. If it was not possible to change the observer, the starting point of the survey and walking direction of the observer were changed. If the *P. apollo* larvae was not detected on either of the two sampling occasions, it does not mean the site was not occupied; only that the species was not detected there.

## 2.4   Data analysing

To estimate the proportion of occupied sites and the probability of site being occupied by the Apollo larvae I used the single species single season occupancy models (MacKenzie et al., 2018c). Shortly, the single-season occupancy analysis focuses on estimating the probability that a site is occupied ($\psi$), along with $p_i$ which is the probability of detecting the species on survey i, given the species is present on the site (MacKenzie et al., 2018a). For the single-season occupancy models, the sites are considered to be closed in the way that the occupancy status of the site cannot change over the course of sampling (MacKenzie et al., 2018a). In this study, non-dispersive *P. apollo* larvae on islands surrounded by the sea is considered to meet the requirements. Since, the single-season occupancy analysis focuses on the different kinds of encounter histories, I used the detection history of the larvae as input data. The detection history of the larvae is a sequence of 1's and 0's indicating whether the species was detected or not (respectively) in each of the surveys of a sampling site.

To examine the possible change in the occupancy of the *P. apollo* larvae I compared the results of the data collected in 2019 to the historical data. I calculated the proportion of occupied sites for naïve occupancy and used occupancy modeling to estimate the probability of site being occupied for the years 1997, 1999-2003 and 2019. Naïve occupancy estimate is the ratio of number of sites where species is detected to total number of sites surveyed, without correcting for imperfect detection (MacKenzie et al., 2018a). I compared both metrics, the naïve occupancy estimates, and occupancy model estimates between the historical data and data collected in 2019. For the historical data that had a record of only a single visit, I corrected the naïve occupancy probability with detection probability of 0.97 based on literature, see Fred & Brommer 2003 for details, to get an estimate of occupancy probability ($\psi$). For the data collected in 2019 I used the encounter history of 44 repeated surveys to correct imperfect detection. In addition, I used constant detection probability (p) in occupancy modeling. I did the comparison of occupancy probability in a smaller scale and in a larger scale between all the survey years. With smaller scale I refer to 58 islands that were surveyed every year in 1997, 1999-2003 and 2019 and a larger scale I refer to all the complete data with all the survey

sites: 1997 (n = 174), 1999 (n = 85), 2000 (n=83), 2001 (n=84), 2002 (n=83), 2003 (n = 74) and 2019 (n = 118).

To test the accuracy of the counting method I studied the relationship of the numbers of *S. telephium* plants of the first survey and second survey occasion. In addition, I performed a single factor ANOVA to test the repeatability of the surveys. I used 36 out of the 44 repeated survey results for the analysis. I excluded 8 of the surveys because of a missing or incomplete value. These 8 excluded surveys were occasions where the people doing a second count did not have time to go through the whole islands edge band. For example, when we had collaborative transportation and the very limited time on survey island was used for detecting possible *P. apollo* larvae instead of accurate count of *S. telephium* plants. Thus, I did not consider these results where plants were counted only in some parts of the island comparable to the first survey and excluded them. However, the successful repeated surveys were largely independent counts given that only 4 out of 36 re-visits the observers were the same in both survey occasions.

To study the possible difference in abundance of *S. telephium* I compared the Sedum classes between data collected in 2019 and the historical data (1997, 1999-2003). By Sedum classes I mean the categories used to score the sites: 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). For the comparison I performed nonparametric Mann-Whitney test that compares two unpaired groups.

To explore if abundance of *S. telephium* affects the occupancy of *P. apollo* larvae I introduced covariates to the general occupancy model. Adding a site covariate to the general occupancy model allowed me to assess whether occupancy of a site is a function of some covariate(s) and could help explain differences in ψ among the sites in the data collected 2019 (MacKenzie et al., 2018a). A site-specific covariate is presumed to affect the probability that a site is occupied, and it should be a relatively stable variable (not likely to change) over the season (MacKenzie et al., 2018a). Therefore, when building an occupancy model with a site covariate, I chose to use *S. telephium* abundance score (Sedum class) as a site-specific covariate because it meets the criteria. In this case, whether a site is occupied or not is a function of the individual site's Sedum class (MacKenzie et al., 2018a). Moreover, because each site has its own Sedum class, each site will end up with a unique probability of occupancy that is directly linked to its corresponding Sedum class (MacKenzie et al., 2018a).

To estimate the detection probability (p) of the larvae in 2019 data I used survey specific covariates in the occupancy modeling. The survey specific covariates could also help explain differences in detection probabilities among the surveys (MacKenzie et al., 2018a). I chose for survey specific covariates the starting time (before noon or

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afternoon), weather (sunny or not sunny) and survey month (May or June). I chose the starting time and weather, because I wanted to examine if there is difference between survey days, and a better probability of detecting larvae when it is warm and sunny. Generally, in the afternoon the air temperature was warmer than before noon, and few mornings before noon were even chilly during the surveys done in 2019. I did not find it reasonable to use more specific time of day than noon since, there were a lot of variation in the survey times between survey days due to weather and transitions from site to another. Furthermore, dividing time covariate to several parts would have complicated the occupancy model excessively. The weather covariate was straightforward to divide in sunny/not sunny covariate because of the larvae sun-basking behavior. The survey month was chosen because I wanted to examine if there were difference in larvae hatching in May and/or June, and if it was possible to determine a difference in probability of detecting larvae in either month. I selected these survey specific covariates because I anticipated that they could help optimize the detection probability in the future surveys.

I noticed that in the data I collected in 2019 there were days when most of the larvae were detected. This could indicate that occupied sites surveyed in the same day and close to each other could have something in common. Therefore, additionally to habitat variable (Sedum class), I wanted to tentatively include a spatial variable to the occupancy models in the 2019 data. To examine the spatial similarity of different survey islands I performed k-means cluster analysis. K-means cluster analysis will randomly locate center points for a given number of centers. I chose 4 as the number of the centers because of the 4 main compass points and 4 Sedum classes in the data. Thus, k-means cluster analysis grouped the survey islands to 4 groups (0-3). After the analysis, to test the possibility that occupancy probability differed across island groups in the study area, I introduced 'island group' as a site-specific covariate for occupancy on the top occupancy model.

I binary coded the collected data for occupancy modelling (Appendix 1). Binary coding is needed for the design matrices that create series of logistic regression equations. Covariate not listed in the matrix was a reference variable that all the other variables are compared to, e.g. I listed sunny weather as a covariate in the matrix and used not sunny weather as a reference. Sedum class 1 and island group 0 were used as references when comparing the site-specific variables.

I started the occupancy modelling from the simplest model and then proceeded to add more variables to the model. I tested each model with constant and survey specific detection probability. The full occupancy model consisted of all possible interactions between the variables. With multiple hypotheses (i.e., occupancy models) about the biological system, I must consider which model (or models) from a set of candidate

models are 'better' for a given data set (MacKenzie et al., 2018c). MacKenzie et al. (2018c) emphasize the use of Akaike's Information Criterion (AIC) approach, since the method can handle non-nested model structures and encourages parsimonious models. Parsimonious models explain the variation in the data well, with as few parameters as possible (MacKenzie et al., 2018c). Thus, as a model selection technique I used information-theoretic method AIC, which rank the models according to a selected metric (MacKenzie et al.,2018c). The magnitude of AIC is not relevant, but the differences in AIC among different models are the focus of model selection (MacKenzie et al., 2018c).

All statistical analyses were performed using SAS Enterprise Guide 7.1. All occupancy models were performed using program PRESENCE 2.12.35 (Hines). The k-means cluster analysis was performed using QGIS 3.4 Madeira. I used the 95% confidence limit in all analyses. Differences at the level of $p \leq 0.05$ were reported as significant or judged by the non-overlapping 95% confidence intervals. I plotted the means with their confidence intervals by using Excel.

## 3  Results

### 3.1  The historical and current occupancy of *P. apollo* in 58 islands

Total of 125 *P. apollo* larvae were detected when collecting data for this study (Table 1). The detected larvae were observed in 22 of all the surveyed islands. There was only one site with more than 20 larvae and 3 sites with more than 10 larvae detected. The weather was mainly favorable for detecting sun basking *P. apollo* larvae, since 52 of 118 surveys had a record of sunny conditions, without wind or clouds. Of these 118 cases, most of the surveys (76 %) were done in May. The average number of *P. apollo* larvae was 1 per site. Of the 118 sites, in 2 sites no counting of *S. telephium* plants was done but the sites were included for further analysis, because of their historical status (surveyed in previous years) and because a larva was detected in both sites in 2019. Only in 2 occasions of all 44 repeated surveys the detection history changed. On one occasion, a larva was detected the first time, but not during the second visit, and on one occasion a larva was detected only when the site was visited the second time.

The hypothesis examining the possible change in occupancy of *P. apollo* larvae in the Archipelago Sea study area was tested at two different levels. First, the occupancy was studied in a smaller scale of 58 islands. Second, the probability of occupancy was examined in a larger scale by including all the historical survey sites, and all sites sampled in 2019 for the analysis. The subset of 58 islands, referred as a smaller scale study, were selected on the basis that the same sites were surveyed each year 1997, 1999-2003 and 2019. Of these 58 islands, 30 sites were visited twice in 2019 for

encounter history. The naïve occupancy estimation and the occupancy model estimates were compared between the historical data (1997, 1999-2003) and data collected 2019 in both scales. The summary of detected larvae in the 58 islands of all survey years is presented in Table 1 with the naïve occupancy estimates for each year.

In the smaller scale examination, the naïve occupancy estimate has been more than 50 % in all historical survey years (1997, 1999-2003) and the estimate was reduced to 5 % in the 2019 survey (Table 1). In addition, the number of detected larvae has decreased by hundreds of individuals. Furthermore, only one of the 58 islands had more than 10 larvae detected in 2019 and before at least 15 % of the islands had over 10 larvae detected.

The smaller scale survey occupancy models resulted in that the estimate of occupancy probability is significantly lower in 2019 than in any historical year as judged by the non-overlapping 95% confidence intervals (Fig. 4).

Both, the general occupancy model estimate of probability of occupancy and the naïve occupancy estimate were 0.05 for the year 2019 (Table 1, Fig.4). Only 3 of 58 surveyed sites was considered occupied since a larva was detected on site at least once. The model and naïve estimates given are the same probably due to small number of sites, few detected larvae and occupied sites, and because the encounter history did not change in any of the 30 repeated surveys included in the model.

There is a clear decrease in the number of larvae, proportion of occupied sites (Table 1), and in the estimated probability of occupancy in the year 2019 compared to the historical survey years (Fig 4).

**Table 1.** Summary of detected *P. apollo* larvae in a smaller scale survey. Every year (1997, 1999-2003 & 2019) the same 58 islands were surveyed. The column "Occupied" is the number of sites where larvae were detected. Column Naïve est. presents the naïve occupancy estimation (Occupied/n). Column No. larvae is the total number of detected larvae. Mean larvae is the mean value of detected larvae per 58 sites. The last column (>10 larvae) is the number of sites where more than 10 larvae were detected.

| Year | n | Occupied | Naïve est. | No. larvae | Mean larvae | >10 larvae |
|------|-----|----------|------------|------------|-------------|------------|
| 2019 | 58 | 3 | 0,05 | 17 | 0,3 | 1 |
| 2003 | 58 | 35 | 0,60 | 683 | 12 | 13 |
| 2002 | 58 | 41 | 0,71 | 313 | 5 | 9 |
| 2001 | 58 | 46 | 0,79 | 737 | 13 | 17 |
| 2000 | 58 | 45 | 0,78 | 587 | 10 | 18 |
| 1999 | 58 | 33 | 0,57 | 259 | 4 | 10 |
| 1997 | 58 | 44 | 0,76 | 438 | 8 | 18 |
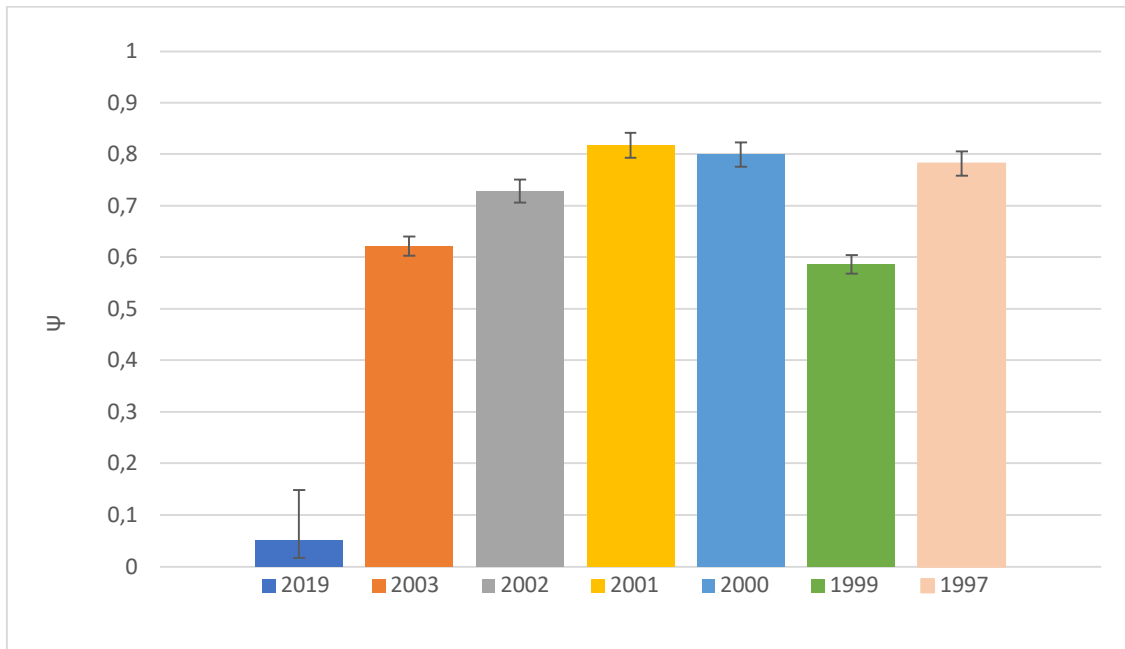
**Figure 4.** The estimated occupancy probability (ψ) of *P. apollo* in a smaller scale of 58 islands. The estimates of occupancy probability with 97 % detection probability for the historical data (years 1997, 1999-2003) and an estimate of occupancy probability with constant detection probability (p) for the year 2019 with 95 % confidence intervals.

## 3.2 The historical and current occupancy of *P. apollo* larger scale survey

The examination of occupancy probability in a larger scale included all historical (1997, 1999-2003) survey islands and all sites surveyed in 2019. In the year 2019 of the 118 sites 44 islands were visited twice for repeated survey. The encounter history of repeated surveys are used to correct imperfect detection and thus, to have more accurate estimation of probability of occupancy. The summary of detected larvae and naïve occupancy estimations for each year are presented in Table 2.

The occupancy models resulted in that the estimated of occupancy probability of *P. apollo* larvae decreased substantially in the year 2019 compared to the historical survey years as judged by the non-overlapping 95% confidence intervals (Fig. 5).

Of the 118 islands surveyed in 2019, *P. apollo* larvae were detected at 22 which leads to a naïve occupancy estimate of 0.19 (Table 2). From the general occupancy model with constant detection probability ψ(.)p(.), the estimated probability of occupancy ψ is 0.22 (Fig. 5). The estimated detection probability (p) based on 118 islands and 44 re-visits was 0.84.

The change in occupancy is clear when comparing the naïve occupancy estimations between historical data and data collected in 2019 (Table 2). Additionally, in the historical data the lowest number of detected larvae were 330 individuals in 1999 which is 205 individuals more than in year 2019. Furthermore, the change in occupancy is clear also

in the occupancy model estimations. Before the lowest estimated probability of occupancy was around 50 % (1997) and in 2019 data the estimation had decreased to circa 20 % (Fig. 5).

The number of detected larvae, the proportion of sites where larvae was detected, and the estimated probability of occupancy have all decreased notably in the past two decades.

**Table 2.** Summary of detected *P. apollo* larvae in all islands. The number of surveyed sites for each year is in column n. Occupied column is the number of sites where larvae were detected. Column Naïve est. presents the naïve occupancy estimation (Occupied/n). Column No. larvae is the total number of detected larvae. Mean larvae is the mean value of detected larvae per surveyed site. The last column (>10 larvae) is the number of sites where more than 10 larvae were detected.

| Year | n | Occupied | Naïve est. | No. larvae | Mean larvae | >10 larvae |
|------|-----|----------|------------|------------|-------------|------------|
| 2019 | 118 | 22 | 0,19 | 125 | 1 | 4 |
| 2003 | 74 | 39 | 0,53 | 708 | 10 | 14 |
| 2002 | 83 | 52 | 0,63 | 338 | 4 | 9 |
| 2001 | 84 | 61 | 0,73 | 792 | 9 | 18 |
| 2000 | 83 | 57 | 0,69 | 678 | 8 | 20 |
| 1999 | 85 | 48 | 0,56 | 330 | 4 | 12 |
| 1997 | 173 | 84 | 0,49 | 591 | 3 | 23 |
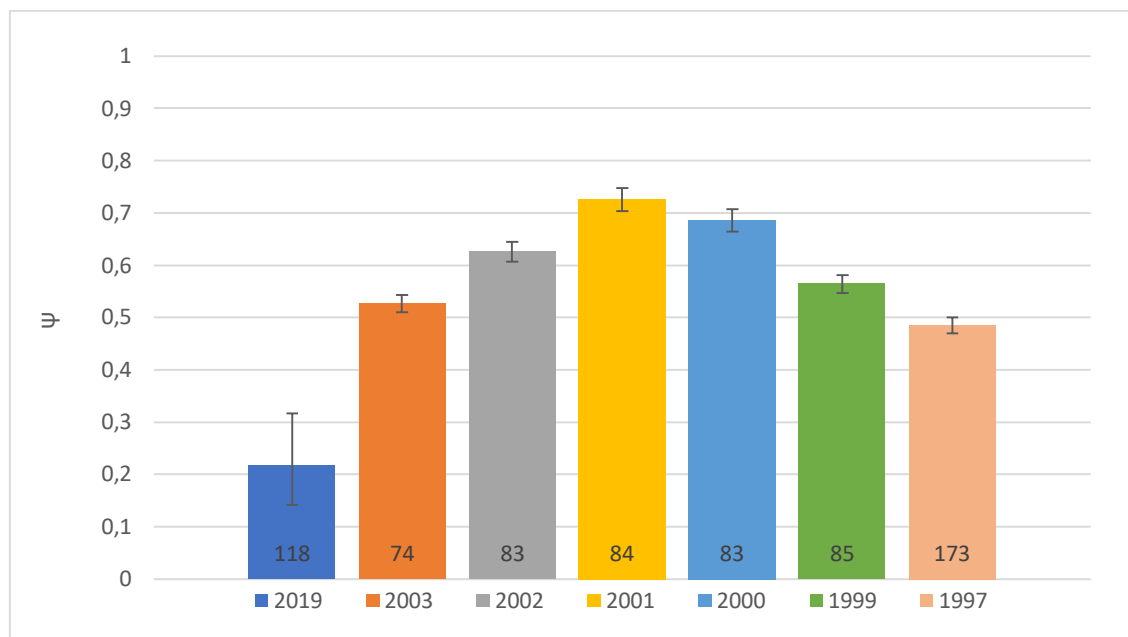


**Figure 5.** Occupancy estimates (ψ) of P. apollo in a larger scale. Estimate of occupancy probability with 97 % detection probability for the historical data (years 1997, 1999-2003) and a model estimate of occupancy probability with constant detection probability (p) for the year 2019 with 95 % confidence intervals for all surveyed islands. Number of survey sites (n) for every year is inside the column.

### 3.3 Repeatability of *Sedum telephium* surveys

In the data collected in 2019 the number of the *S. telephium* plants per site ranged from 0 to 1918. Average number of plants being 214 plants and median value 161 plants. However, in the historical data (survey years 1997, 1999-2003) the number of *S. telephium* plants were scored in categories: Sedum class 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). The same Sedum class categories were used for the data collected in 2019. The class values were used in further analysis.

Of all surveyed sites 44 islands were visited for a second time to repeat the survey in order to test the sampling accuracy. Of these 44 sites, 8 sites with inaccurate *S. telephium* count (observer did not count the whole island) was excluded, and 36 remained for the repeatability analysis. The accuracy and repeatability were good. There was little difference in the counting results of *S. telephium* plants between survey occasions and observers. There is a clear correlation between the two survey results of detected *S. telephium* plants ($R^2$=0.90) (Fig. 6). In addition, I performed the single factor ANOVA to determine the repeatability value. The test resulted in good repeatability value ($F_{35,36}$ = 31.30, p < 0.0001).
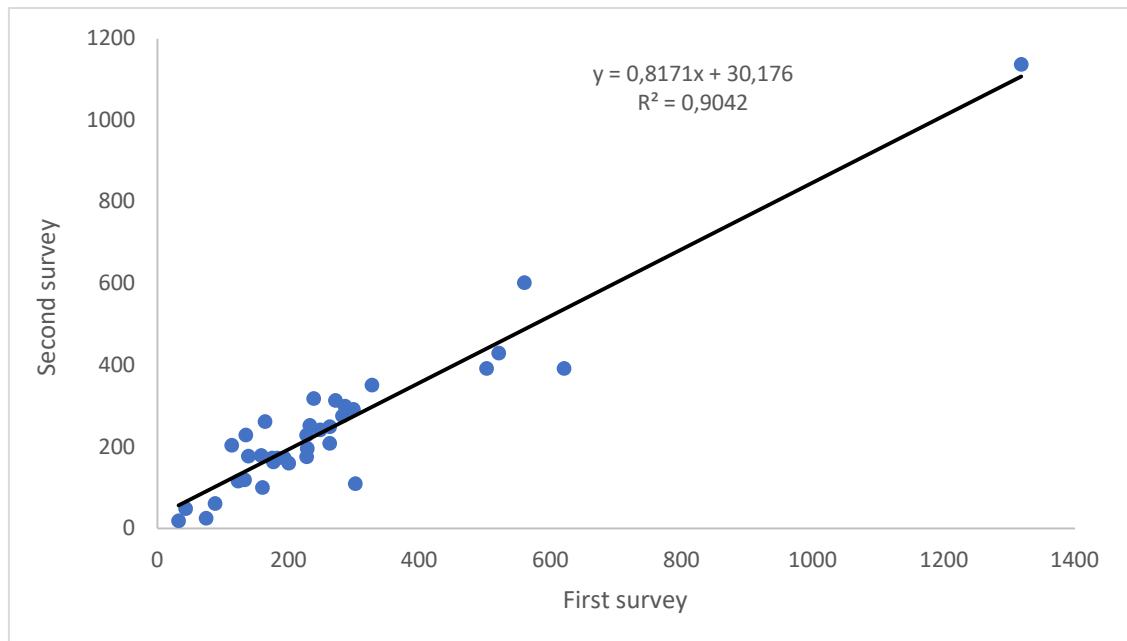


**Figure 6.** The relationship between the number of detected *S. telephium* plants in first and second survey occasions. There is a clear correlation between the two surveys ($R^2$=0.90). Repeated surveys were largely independent, as only 4 out of 36 re-visits the observers were the same in both survey occasions.

## 3.4 The abundance of *Sedum telephium*

The hypothesis exploring the possible decline in abundance of *Sedum telephium* was tested by comparing the Sedum class values (1-4) between the historical data (years 1997, 1999-2003) and the data collected in 2019.

There was no significant difference in the abundance of *Sedum telephium* between historical data and data collected in year 2019 (Table 3, Fig. 7) when comparing the Sedum classes ($F_{1,581}$= 2.14, $p > 0.14$). The overall abundance of *S. telephium* has not decreased in the study area.

**Table 3.** The frequency and percentage of Sedum classes (1-4) over the years. The Sedum class value refers to the number of *S. telephium* plants per site that are scored in categories 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). Historical data was collected in years 1997, 1999-2003 in the same area.

| Sedum class | 2019 | | Historical data | |
|:---:|:---:|:---:|:---:|:---:|
| | n | % | n | % |
| 1 | 4 | 3 | 17 | 4 |
| 2 | 35 | 30 | 176 | 38 |
| 3 | 74 | 64 | 264 | 57 |
| 4 | 3 | 3 | 9 | 2 |
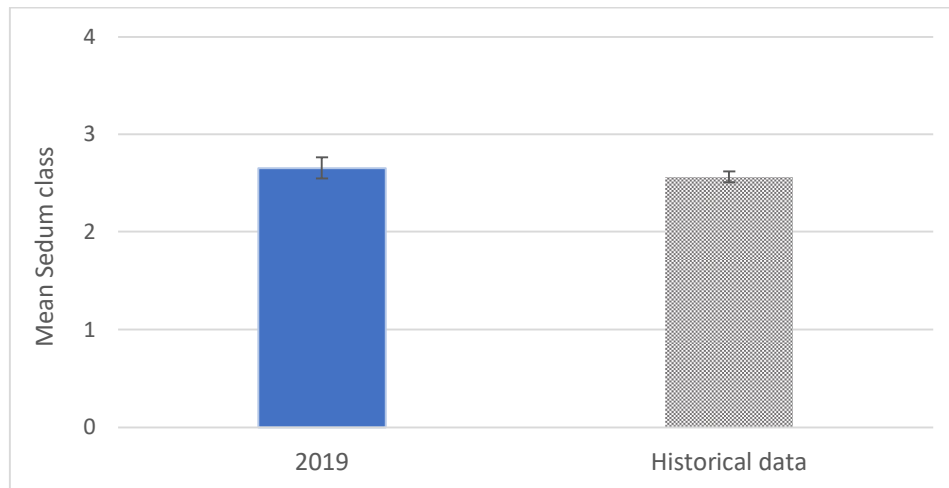


**Figure 7.** The mean values of Sedum class of 2019 and historical data (survey years 1997, 1999-2003) with 95 % confidence intervals. There is no significant difference between the mean values ($F_{1,581}$= 2.14, $p > 0.14$). There is no apparent decline in overall abundance of *Sedum telephium* in the study area.

## 3.5 Factors affecting occupancy and detection probability in 2019 data

Introducing covariates to the general occupancy model allowed me to understand which factors affected occupancy and detection probability in the data I collected in 2019. To assess the differences in probability of occupancy (ψ) among the sites, and to examine if abundance of *S. telephium* affects the occupancy of *P. apollo* larvae, I used the 'Sedum

class' (1-4) as a site-specific covariate. Furthermore, to estimate the detection probability (p), I used survey specific covariates: the starting time (before noon or afternoon), weather (sunny or not sunny) and survey month (May or June). The candidate models were ranked according to their AIC value.

The model with Sedum class as a site-specific covariate for occupancy probability has the greatest support (Table 4). All candidate models in which occupancy is assumed to be constant (.) are at least 9,2 AIC score lower than the most parsimonious model. Thus, the abundance of *S. telephium* may affect the occupancy of *P. apollo* larvae.

The model with constant detection probability, i.e. the probability of detecting the species does not change after the first detection, p(.), has clear support as indicated by the model weights (Table 4). However, there is still some support for model with survey-specific detection probability ψ(.)p(Survey), and candidate models with survey specific covariates on detection. Nevertheless, the top model in terms of AIC is clearly the most parsimonious model with constant detection probability, and the candidate models with covariates on detection are hence more elaborate sub-models of this top model and can be ignored (Arnold, 2010).

**Table 4.** Summary of main occupancy models fit to *P. apollo* data collected in year 2019. ΔAIC is the relative difference in AIC values compared with the top-ranked model; ω is the AIC model weight; Npar is the number of parameters. Models consist of probability of occupancy (ψ) with detection probability (p), and possible covariates included in the model are written inside the brackets. Covariates used are site-specific covariate Sedum class (Sedum), and survey specific covariates starting time (Time), weather (Sunny) and month (May). Covariates not listed act as a reference variable that the other variable is compared to. Time covariate is divided to before noon ad afternoon. Before noon is used as a covariate and afternoon is a reference. In Sedum classes, the class 1 (1-10 plants) is the reference for other classes (2-4). A dot inside brackets (.) indicates that probability of occupancy and/or detection probability is considered to be constant. Survey-specific detection probability is indicated with p(Survey).

| Model | ΔAIC | ω | Npar |
|---|---|---|---|
| ψ(Sedum)p(.) | 0 | 0.208 | 5 |
| ψ(Sedum)p(Sunny) | 1.7 | 0.089 | 6 |
| ψ(Sedum)p(Time) | 1.74 | 0.087 | 6 |
| ψ(Sedum)p(May) | 1.99 | 0.077 | 6 |
| ψ(.)p(.) | 9.2 | 0.002 | 2 |
| ψ(.)p(Survey) | 10.82 | 0.0009 | 3 |
| ψ(.)p(Sunny) | 10.85 | 0.001 | 3 |
| ψ(.)p(Time) | 11.06 | 0.001 | 3 |
| ψ(.)p(May) | 11.19 | 0.001 | 3 |

## 3.6 Occupancy probability as a function of host plant class

The occupancy model with Sedum class as a site-specific covariate for occupancy probability had the greatest support. The model analysis allows to determine whether a site is occupied or not is a function of the individual site's host plant score. The sites were

scored using Sedum class categories 1 (1-10 plants), 2 (10-100 plants), 3 (100-1000 plants) or 4 (> 1000 plants). Each site has its own Sedum class; thus, each site will end up with a unique probability of occupancy that is directly linked to its corresponding Sedum class (Fig. 8).

The results of the occupancy model with Sedum class as a site-specific covariate indicates that the estimate of occupancy probability ($\psi$) is highest in sites scored as Sedum class 2 (10-100 plants) between the surveyed sites. Unexpectedly, the probability of occupancy was higher in Sedum class 2 than in Sedum class 3 as judged by the non-overlapping 95% confidence intervals (Fig. 8). Sedum class 3 have higher abundance of the host plant, thus, it was expected to have higher probability of occupancy based on earlier work highlighting the importance of the host plant abundance (Fred & Brommer, 2003).
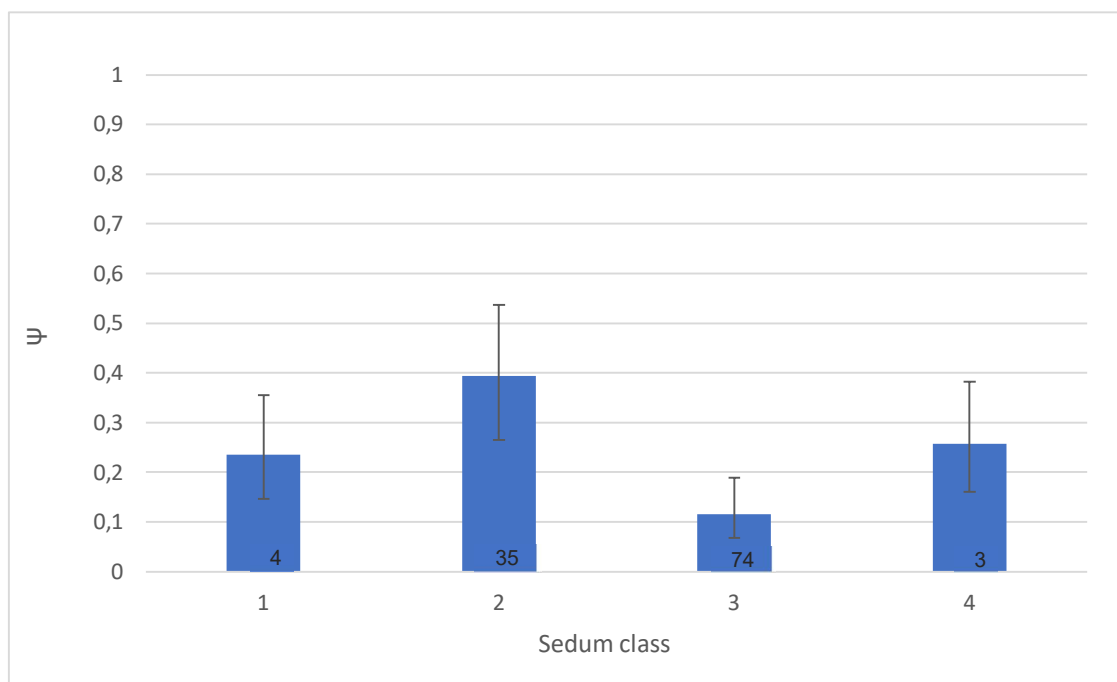


**Figure 8.** Estimate of occupancy probability ($\psi$) as a function of site-specific covariate Sedum class with 95 % confidence intervals. Unexpectedly, the estimate of occupancy probability ($\psi$) is highest in sites scored as Sedum class 2 (10-100 plants). Other Sedum class categories used are: 1 (1-10 plants), 3 (100-1000 plants) or 4 (> 1000 plants). Sample size (n) of each class is inside the column.

## 3.7  Island group analysis

To examine the spatial similarity of different survey islands, the sites were divided into groups with k-means cluster analysis. I chose to use 4 center points for the analysis. The cluster analysis grouped the survey islands according to the spatial location (distance to the center) to 4 island groups or clusters; 0 (n=25), 1 (n=40), 2 (n=32) and 3 (n=21). The

number of islands differed between groupsThe use of 4 center points appears to segment the survey sites as main compass points (Fig. 9).

To test the possibility that occupancy probability differed across island groups in the study area, a site-specific covariate 'island group' was introduced on the top occupancy model (Table 5).

The island group (cluster) as a site-level covariate has the greatest support among the occupancy models, even greater than model with Sedum class as a site covariate (Table 5). This indicates that there is a clear difference between island groups.
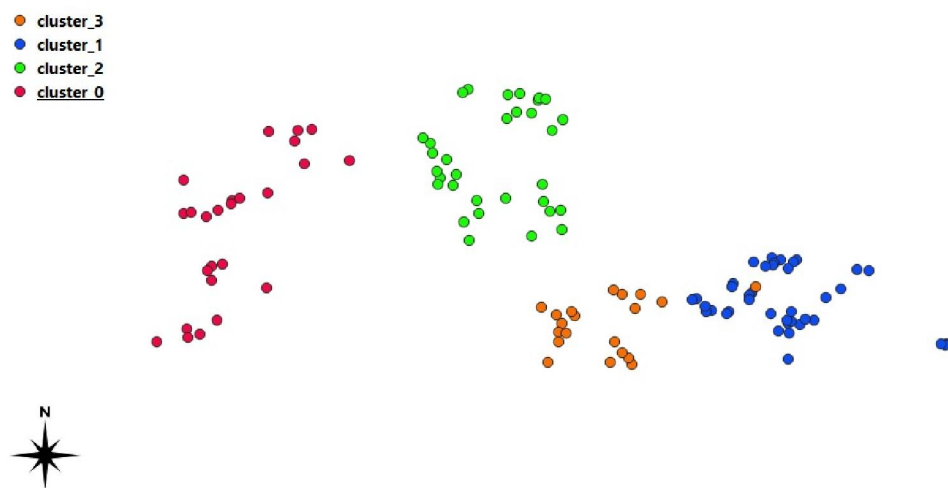


**Figure 9.** The result of k-means cluster analysis**.** Analysis was performed on sites surveyed in the year 2019. The island groups are presented in the survey area with north arrow and without a background map. Different colors indicate different island group. The cluster numbers with colors are presented in upper left corner of the picture. Using 4 center points for the cluster analysis segmented the island groups as main compass points. Island group 0 is in the east (red), 1 in the west (blue), class 2 (green) in the north and class 3 (orange) in the south.

**Table 5.** Summary of top ranked occupancy models fit to *P. apollo* data collected in year 2019. ΔAIC is the relative difference in AIC values compared with the top-ranked model; ω is the AIC model weight; Npar is the number of parameters. Models consist of probability of occupancy (ψ) with detection probability (p), and possible covariates included in the model are written inside the brackets. Covariates used are site-specific covariate island group (Cluster) and/or Sedum class (Sedum). A dot inside brackets (.) indicates that probability of occupancy and/or detection probability is considered to be constant. Survey-specific detection probability is indicated with p(Survey). Island group 0 and Sedum class 1 were used as a reference variables that the other island groups or Sedum class values are compared to.

| Model | ΔAIC | ω | Npar |
|---|---|---|---|
| ψ(Cluster)p(.) | 0 | 0.5258 | 5 |
| ψ((Cluster)p(Survey) | 1.51 | 0.2472 | 6 |
| ψ(Cluster, Sedum)p(.) | 1.68 | 0.2270 | 8 |
| ψ(Sedum)p(.) | 36.71 | 0.0000 | 5 |
| ψ(.)p(.) | 45.91 | 0.0000 | 2 |

# 4 Discussion

In this study, I examined two species, *Parnassius apollo* larvae and their sole host plant *Sedum telephium* in the Archipelago Sea study area. I investigated whether the occupancy of the Apollo larvae has changed, whether the abundance of *S. telephium* has changed and if there is a connection with the possible changes. This was done by comparing data I collected in 2019 to data collected in 1997, 1999-2003 (i.e., historical data) in the same area.

## 4.1 The host plant and larvae

The estimates of occupancy probability of *P. apollo* larvae were compared between historical data and data collected in 2019 to assess whether the occupancy has changed. The investigation of occupancy was done in a smaller scale of 58 islands surveyed every year, and in a larger scale including all the survey islands. The results were that the estimated probability of occupancy has decreased significantly in the study area in 2019 compared to the historical data. Additionally, from the results of smaller scale survey it could be speculated that the 58 islands surveyed every year appear to have lost their importance as breeding habitats for *P. apollo* during the last two decades. In any case, the difference in occupancy was considerable in smaller and in larger scale examinations. The findings are in line with my first hypothesis that the occupancy of the *P. apollo* larvae has changed. In addition to the occupancy model estimates, the number of detected larvae and proportion of occupied survey sites have decreased remarkably in the past two decades. There has been some natural fluctuation in the occupancy of the larvae in previous years. However, in the larger scale survey the number of detected larvae and proportion of occupied sites is less than half in 2019 when compared the previously lowest years (1997 & 1999). In the smaller scale investigation the decline was even more dramatic. Essentially, the results indicate a very strong decline in *P. apollo* population in the study area.

The Sedum class categories (1: 1-10 plants, 2: 10-100 plants, 3: 100-1000 plants & 4: > 1000 plants) were compared between historical data and data collected in 2019 in order to assess whether the abundance of *S. telephium* has changed in the study area. Contrary to the second hypothesis of this study, the overall abundance of *S. telephium* does not appear to have declined. The mean values of Sedum classes used in the comparison show no clear difference between historical data and the data collected in year 2019. It could be speculated that *S. telephium* tolerates possible changes in its habitat better than expected. Nonetheless, *S. telephium* grows on nutrient-poor biotopes that suffer from overgrowth (Hyvärinen et al., 2019). Therefore, it could be just a matter of time before the *S. telephium* patches are covered with grasses and/or other

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vegetation. Regardless, the endangered *P. apollo* larvae depend on their sole host plant. Thus, monitoring and securing *S. telephium* plants in possible *P. apollo* breeding habitats is important in the future as well.

Contrast to my third hypothesis, the abundance of the *P. apollo* larvae does not appear to have declined due to a decrease in abundance of its host plant. Furthermore, most of the detected larvae were found on sites with 10-100 plants instead of sites where host plant is more abundant (>100 plants / site). This result is unexpected, since, according to previous studies the amount and the density of host plant on a patch is the most important habitat parameter and critical factor for *P. apollo* larvae and their survival in the archipelago (Fred & Brommer, 2003). The finding indicates that abundance of *Sedum telephium* is not as important variable affecting occupancy probability of the Apollo larvae as before. The considerable decrease of the larvae albeit their host plant is abundant, is surprising and alarming. It could be argued that the reasons for the decline of the larvae in surveyed sites and in the study area are unknown to this study. However, dividing the survey islands into groups and adding them on top occupancy model resulted in clear difference between island groups. Moreover, the occupied sites were mostly found in the northern part of the study area that may indicate a possible shift of *P. apollo* larvae towards north. The results suggest that the occupancy probability of *P. apollo* larvae is influenced by spatial location. Furthermore, the probability of occupancy may be affected more by spatial location than abundance of the host plant. These findings may aid conservation planning and help direct habitat restoration efforts to appropriate sites.

One critical point is related to the detection probability and the number of repeated surveys. Detection probabilities generally are < 1 (i.e., < 100%), thus a species will often not be detected when it is present (MacKenzie et al., 2018b). MacKenzie et al. (2018b) proposed to correct imperfect detection probability using repeated surveys. Hence, it could be speculated that detection probability of this study, and the reliability of the occupancy model results could have been enhanced with increasing the number of repeated surveys. However, Ewing & Gangloff (2016) presented that repeated surveys may be costly and cause a reduction in the number of sites that can be surveyed. Indeed, the resources were limited, e.g., time, people collecting data and money. Furthermore, the field work in the Archipelago Sea study area greatly depends on the weather conditions and having a way to transit from a site to another. Thus, every repeated survey is a resource taken from finding a novel site possibly occupied by the larvae, which could be considered more valuable information for the CoastNet LIFE project, and for the restoration of habitat patches.

Apart from possible undetected larvae, there are a few critical points more in this study that are essential to acknowledge. First, it can be argued that to what extent the surveyed

sites and the results of occupancy models represent the *P. apollo* larvae actual populations and range. However, the design of this study was to compare the historical data to the collected "current" data. Furthermore, one of the main assumptions of this study was that the historical data include sites that at least previously have served as a stronghold for local *P. apollo* populations. Thus, the possible change in occupancy could be observed based on surveys to those particular islands. Moreover, there has been some natural fluctuation in the occupancy of the *P. apollo* larvae in previous survey years, and it could be argued that the occupancy was merely unusually low in the year 2019 due to dry year in 2018. The drought in 2018 could have affected, e.g., adult or larval food resources thus, could be one explanation for temporary decline. Nevertheless, it is matter of future investigations to examine if additional *P. apollo* populations are found on novel sites, where the occupied sites are, and to what extent there are larvae and occupied sites to be found. However, it is worth noticing that the possibly reported observations of conspicuous and very mobile adult butterfly is not sufficient indicator of the species occupancy, as the presence of larvae is the only reliable sign of a site having been used for breeding (Fred & Brommer, 2005).

As for the accuracy of exhibited decline of the larvae in this study, the change in occupancy is reliable. As mentioned earlier, the change in occupancy was observed when comparing occupancy models estimations between historical data and data collected in 2019. In addition, the change was clear also in the naïve occupancy estimations in the smaller scale and larger scale survey. Ewing & Gangloff (2016) discussed using naïve occupancy for most species to detect population declines. Ewing & Gangloff (2016) suggested that changes in naïve occupancy can be used instead of directly comparing a species' past abundance with current abundance. They continued that measuring a change in naïve occupancy tests the hypothesis that the proportion of sites where a species is detected has changed. Naïve occupancy does not directly test for changes in abundance (Pollock, 2006), but since occupancy and abundance are usually strongly correlated the technique can track population changes (Ewing & Gangloff, 2016). Ewing and Gangloff (2016) added that there are two ways a population can decline. First, population can become locally eliminated from entire sites or second, it may experience declines across its range but not necessarily become eliminated from a site (Pollock, 2006). Moreover, presence-absence data can be used to detect both types of population decline (Ewing & Gangloff, 2016). Additionally, presence-absence surveys are less sensitive to natural fluctuations (Ewing & Gangloff, 2016). Ewing & Gangloff (2016) argued that because presence-absence data can detect situations where a species declines but does not become locally eliminated, it is important not to correct for imperfect detection since this would hide local population declines. However, they also found that in most cases accounting for imperfect detection gives a more

reliable estimation of occupancy. Thus, the results of this study offer several evidences of change in occupancy of *P. apollo* larvae in the study area. In other words, in addition to the occupancy model estimate results, the observed the change also in naïve occupancy estimates indicates severe population decline.

Second, in this study no measurement of quality of the *S. telephium* plants or patches was considered other than abundance. Thus, there might be some other features than abundance of *S. telephium* affecting the relationship between the host plant and the larvae. For example, the individual *S. telephium* plants could be further away from each other's, suffer from drought and/or disease, fast growing grasses could act as a barrier and/or the host plants are otherwise unavailable for the *P. apollo* larvae. However, because of the results of this study, it could be questioned how realistic it is to assume that the change in *P. apollo* larvae occupancy is solely due to the possible changes in host plant patches. Albeit, the earlier work emphasizes the importance of the host plant patches (Fred & Brommer, 2003), and the loss of habitat due to overgrowth of open areas is listed as one of the most significant threats for endangered species living in shores and rocky outcrops (Hyvärinen et al., 2019). As the results from this study were mostly unexpected and differed from the results of previous studies, it is essential that future research examine why the abundance of *S. telephium* does not seem to be as important variable affecting occupancy of the larvae as before

## 4.2   Future research

This study demonstrated a very strong decline of *P. apollo* in Finland. Fred and Brommer are the experts of the subject since they have previously worked for years (1997, 1999-2003) with the Apollo archipelago population collecting data and studying it (Fred & Brommer 2015, 2009, 2005, 2003; Fred et al. 2006; Brommer & Fred 2007, 1999, 1998; Fred 1997). They found no correlation between vegetation cover, slope, orientation, and openness of the border of the area covered by the perennial host plant, and the abundance of Apollo larvae on particular patch (Fred & Brommer, 2003). Therefore, this study concentrated on the abundance of the host plant that was considered the most important variable affecting occupancy of the *P. apollo* larvae (Fred & Brommer, 2010, 2003, 1999). However, the crucial relationship between the larvae and the only host plant was not enough to explain the dramatic decline. Apart from host plant, Fred & Brommer's (2009) research also showed that the adult resources, i.e. nectar plants, are important environmental variable. For instance, in the archipelago population the Apollo females decision to emigrate is affected by the amount of local nectar plants on a patch and the closeness to surrounding nectar resources (Fred & Brommer, 2009). Essentially, the females leave if nectar plants on the patch are limited and take with them the eggs of the next generation (Fred & Brommer, 2009). Therefore, one possible reason for the

population decline is that radical changes in the availability of nectar plant patches have occurred. There very likely are nectar plant patches, but the timing may be disturbed, e.g., the patches flower at the wrong time or maybe they dry out too soon. This would cause the Apollo to abandon the patches of the outer archipelago, even if there is high abundance of host plant. Nevertheless, examining the reason(s) affecting the decline are matter of future research. It is an important matter, as Fred & Brommer (2009) stated that losing even a one patch in the archipelago population would have severe consequenses and the findings of this thesis suggest a loss of several patches. Future reseach is important also for succesful conservation work, as Fred and Brommer (2009) recommended that management planning and operations should be habitat-specific.

There could be several explanations for the decline, e.g., changes in habitat other than abundance of the host plant or not habitat related at all. For example, random factors are listed as one of threats affecting especially species that have very constricted occurrence or small populations (Hyvärinen et al., 2019), such as the Apollo. However, as Sánchez-Bayo & Wyckhuys (2019) presented, one main driver causing the insect decline is climate change. Climate change poses serious and growing threat to many endangered species, since extreme weather events, e.g., drought, rising temperature, changes in ice erosion and loss of snow cover change the living conditions of species living in or near coastlines (Hyvärinen et al., 2019), such as the Apollo. Moreover, van Bergen et al. (2019) concluded in their recent study that the ecological impacts of extreme climatic events on population dynamics and/or community composition are profound and mainly negative. In addition, Kahilainen et al. (2018) pointed out that small metapopulations risk of extinction might essentially increase as extreme weather phenomenon strengthen. Besides the extreme weather events, there are several other potential mechanisms of climate change impacts that may affect on organisms, subpopulations, and thus, species. For example, climate change might change quality in organisms' habitat and microhabitats and/or availability leading to changes in the availability and/or quality of key resources (Foden et al., 2019). In case of Apollo the availability of host plants and/or nectar plants. Furthermore, organisms might experience changes in phenology in a way that the timing of beneficial interactions are disrupted or organisms experience changes in interspecific interactions (Foden et al., 2019). Interspecific interactions include those with beneficial species (e.g., pollinators, dispersers), detrimental species (e.g., competitors, predators, parasite, pathogens) and/or those that are currently neutral but change to beneficial or detrimental in the future. Indeed, while collecting data in the year 2019 I observed some possible interspecific interactions. For example, I discovered multiple species foraging on *S. telephium*. Moreover, in few survey islands I noticed large numbers of generalist, i.e. use many different plants as a larval host plant, *Arctia caja* larvae feeding on *S. telephium*. In addition, I observed a few *S. telephium* plants filled

with ants (*Formicidae*) in some of the survey sites. These are merely couple of examples of probable interactions that may not be beneficial for *P. apollo* larvae. As mentioned before, the reasons for decline of *P. apollo* larvae occupancy might be several, biotic and/or abiotic, and provide need for further studies. Furthermore, it is important to continue to study *P. apollo* as well as other insect species in order to discover proper conservation acts and target them in relevant locations. As the biodiversity of insects is threatened across the globe and the substantial insect declines could trigger wide-ranging cascading effects within many of the world's ecosystems and human welfare (Sánchez-Bayo & Wyckhuys, 2019).

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## Appendices

Appendix 1. The design matrix for program PRESENCE. Sequence of 1's and 0's are indicating whether variable is or is not (respectively), and missing values are indicated with -. In the first four variables (occupancy, time, weather, month) the columns 1. & 2. indicate the first and the second visit to the site.

| site | occupancy 1. | occupancy 2. | Time (AM) 1. | Time (AM) 2. | Weather (Sunny) 1. | Weather (Sunny) 2. | Month (May) 1. | Month (May) 2. | Sedum class (site cov) 2 | Sedum class (site cov) 3 | Sedum class (site cov) 4 | Island group (site cov) 1 | Island group (site cov) 2 | Island group (site cov) 3 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 0 | 0 | 0 |
| 2 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 3 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 4 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 5 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 6 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 |
| 7 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 8 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 9 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 10 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 11 | 0 | 0 | 1 | - | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 12 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 13 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 14 | 0 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 15 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 0 | 0 |
| 16 | 0 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 17 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 18 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 19 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 20 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 21 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 22 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 |
| 23 | 1 | 1 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 0 |
| 24 | 0 | 1 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 0 |
| 25 | 1 | 1 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 0 |
| 26 | 1 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 27 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 28 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 0 | 1 |
| 29 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 30 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 1 |
| 31 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 1 |
| 32 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 33 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 34 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 0 | 1 |
| 35 | 1 | 1 | 1 | 0 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 0 | 0 | 1 |
| 36 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 37 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 38 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 0 | 1 |
| 39 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 | 0 | 1 |
| 40 | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 0 | 1 |
| 41 | 0 | - | 0 | - | 0 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 42 | 0 | - | 0 | - | 0 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 43 | 0 | - | 0 | - | 0 | - | 0 | - | 0 | 0 | 0 | 0 | 0 | 1 |
| 44 | 0 | - | 0 | - | 0 | - | 1 | - | 0 | 0 | 0 | 0 | 0 | 1 |
| 45 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 0 | 0 |
| 46 | 0 | - | 0 | - | 0 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 47 | 1 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 48 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 49 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 50 | 0 | 0 | 1 | 1 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 51 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |

| # | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 52 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 53 | 0 | - | 0 | - | 0 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 54 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 | 0 | 1 |
| 55 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 56 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 57 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 58 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 59 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 60 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 61 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 62 | 0 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 63 | 0 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 64 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 65 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 1 | 0 | 1 | 0 | 1 | 0 | 0 |
| 66 | 0 | 0 | 1 | 0 | 1 | 1 | 1 | 0 | 1 | 0 | 0 | 1 | 0 | 0 |
| 67 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 68 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 69 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 70 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 71 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 72 | 0 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 73 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 74 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 75 | 0 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 76 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 77 | 0 | 0 | 0 | 1 | 1 | 1 | 1 | 0 | 0 | 0 | 1 | 1 | 0 | 0 |
| 78 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 79 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 80 | 0 | 0 | 0 | 0 | 1 | 1 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 81 | 0 | 0 | 0 | 0 | 1 | 1 | 1 | 0 | 0 | 1 | 0 | 1 | 0 | 0 |
| 82 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 83 | 0 | - | 0 | - | 1 | - | 1 | - | 1 | 0 | 0 | 1 | 0 | 0 |
| 84 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 85 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 86 | 1 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 87 | 1 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 88 | 1 | - | 1 | - | 0 | - | 1 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 89 | 1 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 90 | 1 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 91 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 92 | 1 | - | 0 | - | 1 | - | 1 | - | - | - | - | 0 | 1 | 0 |
| 93 | 1 | - | 0 | - | 1 | - | 1 | - | - | - | - | 0 | 1 | 0 |
| 94 | 1 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 0 | 0 |
| 95 | 0 | - | 0 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 0 |
| 96 | 0 | - | 1 | - | 0 | - | 1 | - | 0 | 0 | 0 | 0 | 1 | 0 |
| 97 | 1 | - | 1 | - | 1 | - | 1 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 98 | 0 | - | 1 | - | 1 | - | 1 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 99 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 0 | 1 |
| 100 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 1 | 0 | 1 | 0 | 0 |
| 101 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 102 | 1 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 103 | 1 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 104 | 1 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 105 | 1 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 106 | 1 | - | 1 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 107 | 0 | - | 1 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 108 | 1 | - | 1 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 109 | 0 | - | 1 | - | 1 | - | 0 | - | 0 | 0 | 0 | 0 | 1 | 0 |
| 110 | 1 | - | 1 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 111 | 0 | - | 1 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 112 | 0 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 113 | 0 | - | 0 | - | 1 | - | 0 | - | 0 | 1 | 0 | 0 | 1 | 0 |
| 114 | 1 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 115 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 116 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 117 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |
| 118 | 0 | - | 0 | - | 1 | - | 0 | - | 1 | 0 | 0 | 0 | 1 | 0 |