Captive rearing and translocation of Taylor's checkerspot butterfly (*Euphydryas editha taylori*): South Puget Sound, Washington, 2012-2013

2013 Annual Report to the US Fish and Wildlife Service (Cooperative Agreement F12ACI00835), Joint Base Lewis-McChord Fish and Wildlife Program and JBLM-ACUB Technical Review Committee

17 December 2013

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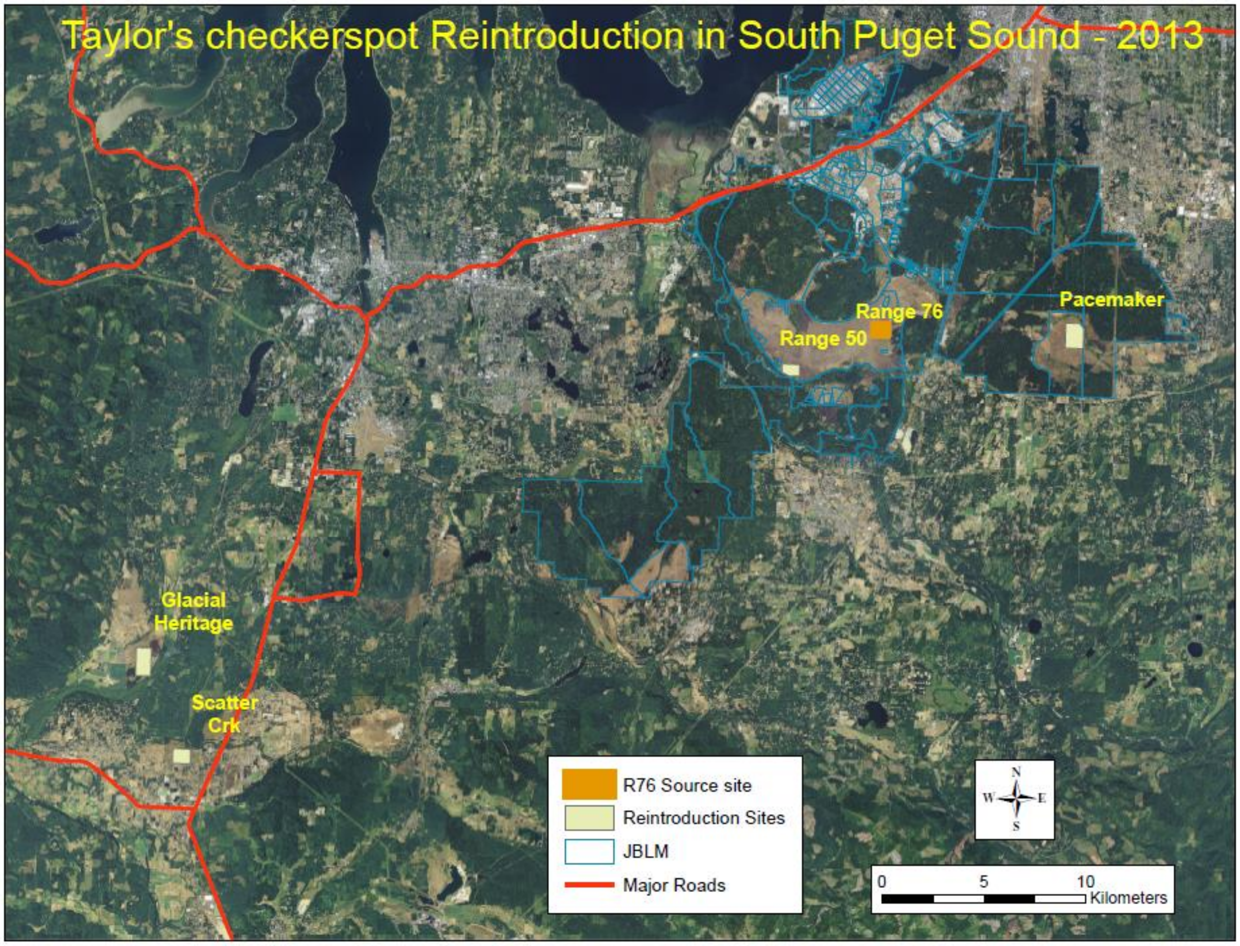
Project Sites

Captive rearing locations are: Oregon Zoo, Portland, OR; Mission Creek Corrections Center for Women, Belfair, WA. Release and monitoring sites (see map below) are: Scatter Creek Wildlife Area – South Unit, (SCS; 2007-2013 releases); Range 50, Joint Base Lewis-McChord (JBLM) (R50; 2009-2011 releases); Pacemaker Airstrip, JBLM (PCM; 2012 release); Glacial Heritage Preserve (GHP; 2012-2013 releases); and Range 76, JBLM (R76; reintroduction source population).



Executive Summary

In 2012, two new sites in South Puget Sound were selected for reintroduction of Taylor's checkerspot: Pacemaker (PCM) and Glacial Heritage Preserve (GHP). Unfortunately Joint Base Lewis-McChord rescinded continued use of Pacemaker for this purpose, so Scatter Creek South (SCS; initiated in 2007), was used as a second release site in 2013 in its place. Range 50 (R50; initiated in 2009) received three sequential releases of Taylor's checkerspot; monitoring continues there and at Pacemaker. An initial assessment of host plant abundance and percent cover of vegetation functional groups for release sites used in 2011 and 2012 suggests that all reintroduction sites appear comparable to occupied plots at Range 76 (R76; extant site), although host plant quantities at GHP in 2012 may have been somewhat lower than desired. A second captive-rearing facility, Mission Creek Corrections Center for Women, began rearing Taylor's checkerspot in 2012 as part of the Sustainable Prisons Project, operated by The Evergreen State College. As of Spring 2013, Mission Creek had reared all life stages of Taylor's checkerspot and successfully mated animals in captivity. As in previous years, most mortality in captivity occurred prior to 3rd instar (Linders 2011b, 2012b), with survival during all remaining stages in the 90th percentiles; this was true for both the Oregon Zoo and Mission Creek. In total 6,589 postdiapause larvae were released on 1 and 4 March 2013. Final dispensation of larvae was altered to include SCS when access to PCM was rescinded by JBLM; the release at GHP continued as scheduled. On 19 May 2013, 107 adults were released at GHP to supplement that population. Post-release monitoring of postdiapause larvae indicated that they were readily observed in all plots at both SCS and GHP until they entered the pupal stage at the end of March. We used distance sampling to quantify daily population density, daily population size, and to illustrate the distribution of adults at the release sites and three other sites, PCM, R76 and R50. In all, 7,397 records were generated across the five sites in 2013, with a total of 3,064 checkerspots counted. This is just one third of the number of checkerspots counted during the 2012 flight season. Checkerspots were present in throughout the sampling areas at both R50 and R76, whereas distributions at SCS, and GHP tracked the areas of release more closely. Adult counts were lower than expected based on postdiapause larval surveys and were also reduced on sites where no releases occurred. More work is needed to associate weather data and characteristics of flight season length, timing and stage-specific Taylor's checkerspot abundance, however monitoring of both postdiapause and adult stages is helpful in clarifying this relationship. Long-term monitoring and population goals were developed in fall 2012. Based on those criteria, R50 far exceeded the target of 250 adults on a single day on the first year in which the population was based solely on reproduction, with a peak single day abundance estimate of 2,058 adults (range 1,467-2,886) in 2012. Raw counts for 2013 (Table 16) suggest abundance estimates will also exceed the target for 2013. In addition adults were distributed across the nearly the entire 22-ha monitoring area in both 2012 and 2013. Baseline standards will need to be met for five consecutive years based solely on reproduction in the field to meet the actual threshold established for success. This report summarizes captive-rearing work conducted in May 2012-May 2013, and reintroduction work from July 2012-July 2013; several cross-year analyses are also included.

Project Goals and Objectives

The goal of this project is to establish new Taylor's checkerspot populations in Washington's South Puget Sound to reduce the likelihood of local extinction, and eventually move toward species recovery. To accomplish this, WDFW and its partners intend to establish at least three new populations at three sites in the next decade. This project employs a strategy of captive rearing and reintroduction which has been funded jointly by JBLM's ACUB program and the USFWS Recovery Initiatives Program, with in-kind support from JBLM, the Oregon Zoo, The Washington State Department of Transportation Habitat Enhancement fund, the Washington Department of Corrections, and ACUB cooperators. This report mainly summarizes activities from the 2012-2013 field season, although some longer term summaries are also included. In 2013 we proposed to 1) release a total of about 4,000 postdiapause larvae at 4 reintroduction sites, 2) conduct follow-up surveys during postdiapause larval, and adult life stages at reintroduction sites, 3) conduct prediapause larval surveys at one reintroduction site, and 4) conduct surveys of adult butterflies at one additional reintroduction site. To insure clarity and cohesion, this report covers all activities involved in captive propagation and reintroduction of Taylor's checkerspots in 2012-2013, regardless of funding source.

This project has five main objectives, which also form the structural framework of this report. Some objectives include a suite of smaller tasks and objectives, which are emphasized by additional headings in the text. The objectives are:

I. Select sites for reintroduction of Taylor's checkerspot in South Puget Sound

II. Produce larvae for release via captive propagation and collection of wild stock

III. Release captive and associated wild stock

IV. Monitor success of the reintroductions

V. Long-term monitoring and population goals

I. Select sites for reintroduction of Taylor's checkerspot in South Puget Sound

Methods

A suite of historic and potential sites within the known range of Taylor's checkerspot in South Puget Sound were initially scored in 2006 (Linders 2006), however restoration trajectories since 2006 have varied by site as a result of restoration priorities, funding source, competing land uses, and cross-species conservation efforts. The objective of Taylor's checkerspot habitat restoration in South Puget Sound has been to return degraded grasslands to a forb-rich condition that can be readily characterized as containing dense and diverse host and nectar plants with a low, open vegetation structure. Priority host species include the perennials *Plantago lanceolata* and *Castilleja hispida*; priority annuals include *Collinsia parviflora* and *Plectritis congesta*. Key nectar sources include *Balsamorhiza deltoidea*, *Armeria maritima*, *Lomatium triternatum*, *L. utriculatum*, *Fragaria virginiana*, *Camassia quamash*, *Saxifrage integrifolia* and *Plectritis congesta* among others (Jackson 1982, Linders 2006, Linders 2011b, Linders 2012b). Ideally, the tallest plants during the flight season will be forbs such as *Balsamorhiza*, *Lupinus alba*, and *Lomatium* to insure access to nectar, basking and roosting sites, and oviposition locations. On sites with an open vegetation structure the ground surface is readily visible even if plants and other organic materials (e.g., moss, lichen, rock, litter, etc.) are present.

Habitat conditions on potential reintroduction sites were periodically reviewed by project partners via annual field visits, with final site selection occurring on an as-needed basis. Factors considered in selection of reintroduction sites included historical checkerspot presence, host and nectar plant abundance, habitat condition (vegetation density, structure and diversity), land owner willingness and funding source (ACUB funds were for management off JBLM; USFWS funds were not spatially restricted).

Select areas for release of larvae

Once a release site is chosen, release areas are selected based on ocular assessment of host plant abundance, including multiple host plant patches, each at least 4 square meters in size with an average of at least 10 perennial host plants per square meter (Linders et al. 2009, Severns and Warren 2008, Severns and Grosboll 2011, Grosboll 2011). Host plant patches typically consist of *Plantago* over-planted with other host species (*Plectritis congesta*, *Collinsia parviflora*), although patches of *Castilleja* also typically occur. One *Plantago* plant (10 x 10 x 10 cm) per released larva is the target minimum based on feeding requirements at the Oregon Zoo. This strategy helps ensure sufficient and diverse food resources throughout larval growth periods. Secondary consideration is given to nectar abundance and diversity, and areas with a low, open vegetation condition. In the year prior to release Mary Linders works with the land manager to identify any outstanding restoration actions, such as increasing local host diversity by seeding annuals. Alternatively they may be asked to do some additional spot-spraying or removal of small broom to improve the low, open condition of the habitat for basking. A site may be deemed suitable to begin receiving larvae when at least 1,200 square meters of habitat dominated by host plants (enough for two release plots) and at least 12,000 square meters (about 3 acres) of supporting habitat (fewer host resources but otherwise as defined above) have been prepared. These habitat amounts originate from field observation at Range 76, where postdiapause larvae have often been observed at a density of 1 larva per square meter and an approximation of the spread of adults in the first year following release.

Obtain measures of habitat condition and host plant availability during the postdiapause larval feeding period to compare between occupied and unoccupied sites.

To assess the condition of postdiapause larval release plots relative to similarly occupied locations at Range 76, we measured relative cover of vegetation functional groups as well as host plant density and frequency by species at R76 in 2012 and in release plots at reintroduction sites in 2009-2012. To select plots at R76 we used a high-resolution aerial photo flown in 2011 and a mapping grid showing *Plantago* distribution (Lyon 2007) to identify two 100 x 125–m areas in ArcGIS where postdiapause Taylor's checkerspot larvae have been observed annually (Linders, pers. obs.). Each 100 x 125–m area was divided in half to distribute samples across the blocks, and a series of corner points (SW corner of 25 x 25-m cells used to map vegetation) were chosen randomly from each half. On one occasion, a cell in the field contained only a few scattered host plants, so it was discarded and the next random cell used instead. A total of three non-overlapping 25 x 30-m plots were established and surveyed as a result. Sampling at R76 occurred when there was sufficient sun to cast shadows and temperatures were > 50° F, which minimized impacts to larvae and allowed us to verify plot occupancy.

To systematically distribute effort and avoid overlapping samples, each 25 x 30-m plot (actual plot sizes vary by site and year) was subdivided into 5 x 5-m subplots. One 0.5 x 1.0-m sampling plot was located 0.5 m west and 1.0 m north of the southeast corner of each 5 x 5-m subplot. Analysis of previous data suggested that increasing the number of 0.5 x 1.0-m sampling plots by 25 percent led to a more stable mean (Linders, unpub. data). So beginning in 2012, seven randomly chosen subplots received a second sampling plot placed 0.5 m east and 1.0 m south of the NW corner of the sub-plot.

Vegetation functional groups and host plant abundance (Table 1) were assessed in all plots in late-February (i.e., about half way through the postdiapause larval feeding period). Reintroduction plots were assessed prior to the release of larvae; larvae were present in all plots sampled at R76. A cross-hair point intercept frame with 36 intersections was used to collect percent cover of different functional groups within each 0.5 x 1.0-m sampling plot. A 0.5 x 1.0-m PVC frame was placed in the same location to obtain host plant counts by species. All host plants with centers inside the frame were counted as well as host plants whose centers fell along one long and one short side of the frame. One *Plantago* or *Castilleja* plant was defined as all those rosettes or stems, respectively, which appeared to originate from a common center. Data were entered on standardized forms and transferred to an Excel data base.

Table 1. Categories of functional vegetation groups and host plant species used to collect percent cover and frequency data at extant and reintroduction sites for Taylor's checkerspot in South Puget Sound, Washington 2009-2012.

Percent cover categories
PLLA: *Plantago lanceolata*
CAHI: *Castilleja hispida*
COPA: *Collinsia parviflora*
PLCO: *Plectritis congesta*
FORB: Any forb
FERN: Any fern; bracken fern is most common
MOSS: Any moss
FESC: Any fescue
GRAS: Any grass
BARE: Bare ground, rock, or lichen
THAT: Thatch, dead material that is <u>disconnected</u> from the plant
WOOD: Any woody material, live or dead. Includes live shrubs
TRAS: Trash, metal, plastic, etc.

Frequency categories
of *Plantago lanceolata*
of *Castilleja hispida*
of *Collinsia parviflora*
of *Plectritis congesta*

Results and Discussion

Four sites have been selected for reintroduction to date from the larger list of potential sites in the South Puget lowlands. Two sites, SCS (since 2007), and R50 (since 2009) have received multiple sequential releases of Taylor's checkerspot (Appendix A). Two additional sites, (PCM and GHP), were initiated in 2012; sequential releases were expected to continue for five years. Unfortunately JBLM denied access to continue releases at PCM in 2013, although the site will continue to be monitored.

Select areas for release of larvae

Prior to release of checkerspot larvae, reintroduction sites received a number of treatments designed to enhance habitat and increase checkerspot survival. Sites were initially treated for invasive shrubs, grasses and forbs using one or more of the following treatments: prescribed fire, mowing, brush-cutting, and repeated applications of broad-spectrum and grass-specific herbicides (e.g., see Fimbel and Dunn 2012). Entire sites have not yet been restored, so release plots for Taylor's checkerspots were initially identified based on an ocular assessment of host plant abundance, with secondary consideration given to nectar abundance and diversity, and a low, open vegetation structure. Areas with more open ground, lower grass and moss density, and at least a background scattering of host and nectar plants were selected. Host plants were added to release plots at a minimum of one mature (10 x 10-cm) plant per larva, with host plants composed primarily of *Plantago lanceolata*. This host plant density was based on feeding requirements observed at the Oregon Zoo. While released larvae have also been observed feeding on *Castilleja hispida* and *Collinsia parviflora*, all lowland populations of Taylor's checkerspot in Washington and Oregon currently depend on *Plantago* as their main or only host (Severns and Warren 2008, Severns and Grosboll 2011). Release areas (release plots and vicinity) at SCS, R50, PCM and GHP were also enhanced with seeds of the larval hosts *Plectritis congesta* and *Collinsia parviflora*; *C. hispida* seed was used at all sites except PCM; *P. lanceolata* seed was used only at GHP and SCS. This strategy was employed to ensure sufficient and diverse food resources throughout larval growth periods, which has the potential to increase larval survival, especially in the face of climatic fluctuations (Hellmann 2002). Native seed resources became available in quantity for the first time in 2011, and production still lagged behind the quantities required by land managers for restoration in 2012. Plugs of *C. hispida* were also added in and around release plots at SCS, PCM and GHP, as well as plugs and seed of suitable nectar species (e.g., *Balsamorhiza deltoidea*, *Armeria maritima*, *Lomatium* spp., *Fragaria virginiana* and others). Adult butterflies are attracted to nectar, which can increase egg production and longevity of adults, and increases the likelihood that females will oviposit in the area (Murphy 1983, Murphy et al. 1983). Only seeds of native host and nectar plants were used at R50, as digging is not permitted and *Plantago* is already widespread and abundant (Lyon et al. 2007). Ultimately, habitat treatments varied by site based on existing host, nectar and vegetation densities, availability of seeds and plugs, the amount of open ground, and the timing and character of prescribed fire (see Fimbel and Dunn 2012).

Release areas were enlarged and made amorphous (as opposed to rectangular) in 2013 to accommodate an increased number of larvae available for release and to take better advantage of restored areas. Three large plots and six plots of varying size were available for release at SCS and GHP, respectively. Sizes of individual plots as well as total release areas are shown in Table 2.

Table 2. Size of individual release plots and total area available for release of Taylor's checkerspot larvae at Scatter Creek South and Glacial Heritage, South Puget Sound, Washington, Spring 2013.

Site/plot	Hectares	Acres
Scatter Creek South – total	0.302	0.746
Plot H	0.185	0.457
Plot I	0.025	0.062
Plot J	0.092	0.227
Glacial Heritage Preserve	0.227	0.619
Plot A	0.100	0.247
Plot B	0.031	0.077
Plot C	0.060	0.149
Plot D	0.014	0.035
Plot E	0.022	0.054
Plot F	0.023	0.057

Obtain measures of habitat condition and host plant availability during the postdiapause larval feeding period to compare between occupied and unoccupied sites.

Percent cover of functional vegetation groups and frequency of host plants by species are summarized by site for occupied plots at R76 (extant site) and prepared release plots at reintroduction sites (R50, PCM, GHP) for 2011-2012 (Table 3). Data collected in 2009 and 2010are not presented as they are based on fewer plots samples and more work is needed to determine whether they are comparable. Differences between sites were more distinctive than differences between plots. As expected, the amount of host plant varied at both the plot scale and at the site scale, however all reintroduction sites appear to compare favorably with occupied plots at R76 in both frequency and cover of host plants. More grass on the 91st Division Prairie sites (R76 and R50) relative to SCS, PCM and GHP is one of the more notable and perhaps unexpected differences (Table 3). The restriction of fern and wood to plots at SCS are consistent with the deeper, moisture-rich soils that characterize that site. Plots at SCS in 2011 and GHP in 2012 were over-seeded with *Collinsia parviflora* and *Plectritis congesta*; 2012 seed available for these species at PCM was used in a single dense planting block that was part of a third release plot for which vegetation data were not available. *P. congesta* found at PCM was the result of a previous seeding.

II. Produce larvae for release via captive propagation and collection of wild stock

Our objective with respect to captive propagation is to produce the target numbers of eggs, larvae and adults for reintroductions proposed in Appendix A. Captive propagation methods were developed at the Oregon Zoo (Barclay et al. 2009) and have been transferred to a second rearing facility housed at Mission Creek Corrections Center for Women in Belfair, Washington and operated via the Sustainability in Prisons Program at The Evergreen State College. More details on the design, construction and development of the program can be found in Linders (2011c).

Continue captive propagation at the Oregon Zoo and Mission Creek to achieve target numbers of eggs, larvae and adults for reintroductions

Methods

Collection of wild females

A population size of at least 1000 adults at Range 76 is sufficient to supply 20 females (not to exceed 2 percent of the local population) for oviposition. Should something happen to the wild population, 10 females per rearing facility annually is sufficient to sustain a captive population based on guidelines provided by the Population Management Center (Schad 2008). Wild females are collected in the hopes that they will supply two thirds of the 10,000 eggs needed, with the remaining eggs (n = 3330) supplied by captive-mated females. This is double the number that was targeted prior to the addition of the Mission Creek facility in 2012.

The large population at R76 allowed us to collect 24 wild females (11 to Oregon Zoo and 5 to Mission Creek on 19 May; and 8 to Mission Creek on 24 May) for oviposition in 2012, to expand the size of the 2012-2013 captive-rearing founder population and reduce the potential influence of captive propagation on stock used for reintroduction. Several alternative strategies to collection have been identified (see Linders 2011a) in the event numbers in the wild are lower than desired. Females showing light wing wear and with swollen abdomens are targeted for collection from locations across the site to maximize genetic diversity and minimize impacts to the source population. Soft nets are used if necessary to capture adults and handling is minimized to reduce injury. Temperatures were relatively cool the morning of 24 May, which kept adults grounded on their overnight roost sites. This allowed us to visually inspect a few hundred females prior to collecting the freshest looking females. However there was relatively little difference in condition among them, and all appeared somewhat emaciated. A spring drought the first half of May in 2012 with temperatures >26 °C (80 °F) eliminated flowers on most plants and is thought to have contributed to the poor condition of wild adults. Adult butterflies were transferred to clean insect jars and labeled numerically with the date and collection location, then transported to the Oregon Zoo and Mission Creek in coolers.

Captive mating

Offspring from 21 maternal lines were available for inclusion in the 2012 captive breeding colony. This was more than the Oregon Zoo could manage, and while Mission Creek was available to assist, this was their first attempt at mating adults. To take advantage of the expanded capacity while managing risk, 7 matrilines were retained at the Oregon Zoo and 5 matrilines were sent to Mission Creek to initiate mating trials there (Fig. 1); three matrilines did not contain enough offspring to be included in the breeding colony. Thirty larvae from each wild matriline are typically held in captivity for breeding purposes. In 2012, lines containing at least 55 larvae were used for breeding at Mission Creek, which allowed an additional 25 larvae from those lines to be held at the Oregon Zoo as "back-up" breeders in the event a problem occurred at Mission Creek. Adults that were strong enough to eat and move freely were assigned a composite number incorporating year of origin, year eclosed, female lineage, and individual ID, and included as breeding stock at the Oregon Zoo. Pairings were determined using a "daisy chain" approach (Fig. 1) based on guidance from the American Zoological Association's Population Management Center at the Lincoln Park Zoo, Chicago, Illinois (e.g., see Lewis et al. 2013, *in prep*.). Our objective was to obtain two successful copulations from each dyad in the daisy chain. Mating was achieved by co-locating a female with a group of males; all copulations and mating attempts were documented (Barclay et al. 2009). Copulating pairs were housed separately and labeled with individual and ancestral lineage ID, weather conditions and time copulation began. Cups were placed near the window and checked frequently to note the separation time. Following separation, the male

Table 3. Mean percent cover and standard deviation (SD) of larval host plants (PLLA, PLCO, COPA, CAHI) and functional plant groups (top) in occupied plots at an extant site (R76; 2012) and in prepared plots at reintroduction sites (SCS and R50 – 2011 and 2012; PCM and GHP - 2012) in February, just prior to release of postdiapause Taylor's checkerspot larvae in South Puget Sound, Washington. Mean frequency (#/m^2) and standard deviation (SD) of host plants (bottom) are also shown. R76 = Range 76; R50 = Range 50; PCM = Pacemaker; GHP = Glacial Heritage Preserve; PLLA = *Plantago lanceolata*; PLCO = *Plectritis congesta*; COPA = *Collinsia parviflora*; CAHI – *Castilleja hispida*; FORB = all non-host forbs; FESC = *Festuca* spp.; GRAS = other graminoids; FERN = any fern; WOOD = dead wood or shrub; THAT = loose thatch; MOSS = any moss; BARE = bare ground, lichen and rock. Number of plots and 5 x 5-m subplots (see text for details) for 2011: SCS = 2/48, R50 = 2/48; for 2012: R76 = 3/111; R50 = 1/44; PCM = 2/74; GHP = 3/77.

	2011				**2012**							
	SCS		**R50**		**R76**		**R50**		**PCM**		**GHP**	
Group	**% cover**	**SD**	**% cover**	**SD**	**% cover**	**SD**	**% cover**	**SD**	**% cover**	**SD**	**% cover**	**SD**
PLLA	2.1	0.2	2.3	1.1	3.3	1.9	4.7	na	5.4	2.2	2.1	0.8
PLCO	1.1	0.2	0.0	0.0	0.0	0.0	0.0	na	0.0	0.0	0.3	0.5
COPA	5.7	1.0	0.0	0.0	0.0	0.0	0.0	na	0.0	0.0	0.1	0.2
CAHI	0.0	0.0	0.0	0.0	0.0	0.0	0.0	na	0.0	0.1	0.0	0.0
FORB	12.9	2.7	12.2	1.6	13.2	1.7	16.7	na	12.7	2.4	18.1	5.8
FESC	4.7	5.3	40.2	4.2	12.7	11.8	2.0	na	9.0	3.8	2.1	1.9
GRAS	12.8	2.2	25.2	10.4	30.1	16.0	34.6	na	13.5	6.3	13.1	6.1
FERN	1.8	0.1	0.0	0.0	0.0	0.0	0.0	na	0.0	0.0	0.0	0.0
WOOD	0.9	0.1	0.0	0.0	0.0	0.0	0.0	na	0.0	0.0	0.0	0.0
THAT	7.7	0.9	4.4	3.8	9.7	0.1	8.1	na	33.9	11.5	17.4	7.1
MOSS	14.7	5.9	9.5	0.3	17.7	7.6	19.9	na	19.6	2.0	28.2	9.5
BARE	35.5	9.7	6.3	0.0	13.2	3.6	13.9	na	5.9	3.3	18.6	2.8
Host	**#/m2**	**SD**	**#/m2**	**SD**	**#/m2**	**SD**	**#/m2**	**SD**	**#/m2**	**SD**	**#/m2**	**SD**
PLLA	5.5	0.2	3.5	2.7	7.9	3.1	13.9	na	22.4	10.6	7.2	4.3
PLCO	37.6	11.5	0.0	0.0	0.0	0.0	0.0	na	0.4	0.6	11.6	14.1
COPA	150.0	29.1	0.0	0.0	0.0	0.0	0.0	na	0.0	0.0	2.5	3.3
CAHI	0.0	0.0	0.0	0.0	0.0	0.0	0.0	na	0.5	0.4	0.6	0.2

was placed in a separate tent with other males that had already copulated. Limiting copulations to one per male is done to increase the genetic contribution from each line. The female was fed and placed in an oviposition chamber, unless no copulation occurred, in which case she was returned to the refrigerator. All males are used in the mating process because they benefit from being housed in a group, however only a few females from each line are needed for egg production; remaining females are released.

Captive rearing, egg to adult

Taylor's checkerspot readily oviposits in captivity. Our goal was to collect about 150 eggs from each gravid wild and captive female to meet a target of 5,000 eggs at the Oregon Zoo in 2012. Because Mission Creek was only just beginning to work with Taylor's checkerspot, we did not set specific goals for egg production, although we did anticipate they would be able to produce 1200 postdiapause larvae. Oviposition and rearing methods followed Barclay et al. (2009); more detail on captive propagation methods used in 2012-2013 can be found in Lewis et al. (2012). Eggs and larvae were checked daily and hatch date recorded. Larval numbers cannot be assessed accurately until 3rd instar, when they are hardy enough to be manipulated individually; at this point an official "hatch" count is obtained. Both pre- and postdiapause larvae were reared exclusively on freshly-cut *Plantago lanceolata* leaves rather than on native host plants because it is 1) easy to grow and handle in the lab, 2) it is less prone to mold and desiccation, and 3) results in high survival (Linders 2007). Oviposition host plant choice in adult females is a genetically derived trait that is not affected by the host plant on which they fed as larvae (Singer 2004).

Once larvae entered diapause no further feeding or handling occurred. We refer to the time period from diapause initiation until the time the weather cools and larvae are moved outdoors as "warm" diapause. Subsequently, larvae were moved outdoors for the remainder of the "cold" diapause period. Rearing cups were placed inside inverted clay pots which are seated inside a clay saucer; pots rest on wood pallets underneath a building overhang at the Oregon Zoo. Larvae were checked periodically during diapause and dead animals removed. Because testing of a suitable diapause location was still underway at Mission Creek, most larvae were transferred from there to the Oregon Zoo on 22 August 2012 for overwintering. Mission Creek retained 450 larvae to test their own diapause location in the small room of the greenhouse.

Larvae were removed from diapause in late February 2013 following increased activity levels, sunny weather and emergence of wild progeny. A total of 3,106 diapausing larvae were transferred back to Mission Creek to be removed from diapause there. The balance of the larvae remained at the Oregon Zoo. Upon removal from diapause, larvae were placed in a high humidity environment and provided with fresh food (Barclay et al. 2009). Once they began to eat, a subset of larvae from wild females captured in 2012 was retained at each facility for inclusion in the 2013 breeding colony; the remainder, including all offspring of captive-mated females, was released.

Larval rearing locations at the Oregon Zoo varied based on purpose (release vs. breeding) (Lewis et al. 2013, *in prep*.). Larvae destined for release were held in indoors for only a short time to maximize development time in the wild and reduce the likelihood that they would reenter diapause. Larvae destined for captive breeding at the Oregon Zoo were reared outdoors in a screen tent, which increases the likelihood that they develop on a similar timeline to those in the wild and has been found to produce larger adults (Linders 2010). Pupae were brought indoors to complete development, while larvae returning to diapause were brought indoors and retained as in the past (Barclay et al. 2009). All larvae at Mission Creek were reared in the greenhouse regardless of purpose and destination. Rearing of larvae, pupae and adults at both facilities followed standard protocols (Barclay et al. 2009).

Rearing conditions

During all stages of larval growth at the Oregon Zoo, full spectrum fluorescent lights were maintained on a 12-hour light/12-hour dark cycle from 0700-1900 using timers. Indoors, overhead lights were turned on during the day to increase light intensity. Target conditions for temperature, relative humidity, and supplemental heat and light are summarized by life stage in Table 4. To reduce wing wear and improve condition of males, heat lamps were turned on for two 30 minute blocks (Table 4), rather than the 4-h block used previously (Linders 2012b). Conditions during mating were generally the same as for adult males, but mating tents were also placed next to a window or outside on a sunny day. Oviposition chambers were placed outside on sunny days. Temperature and relative humidity during all rearing stages were measured as min/max data downloaded at the same time each day (1100-1300 h).

All rearing at Mission Creek was done in the main 16 x 10-ft room of a glass greenhouse; a secondary room 8 x 10-ft room was used to provide early season plants and for mating purposes. Target conditions for temperature and relative humidity were the same as at the Oregon Zoo (Table 4); no supplemental light was provided.

Table 4. Target conditions for temperature and relative humidity, and provision of supplemental heat and light, summarized by life stage for Taylor's checkerspot in captivity, Oregon Zoo, Portland, OR and Mission Creek Corrections Center, Belfair, WA, 2012-2013.

Life stage	Temp (°F)	RH (%)	Supplemental Heat/Light
Males	ambient	ambient	160-watt mercury vapor lamps 30 min @ 1000 & 1400 h
Oviposition	ambient	ambient	160-watt mercury vapor lamps 30 min @ 1000 & 1400 h
Egg/prediapause	65-68	50	
Warm diapause	ambient	ambient	
Cold diapause	ambient	ambient	
Postdiapause	<65	≥55	50-watt tungsten lamps at 1000-1400 h
Pupation	>65	>50	

Results and Discussion

Captive mating

The Oregon Zoo conducted 16 planned breeding introductions among the primary breeders, resulting in 14 copulations, an 87.5 percent copulation success rate and a significant improvement in efficiency over previous years. The first copulation occurred on 10 May 2012, and the last on 19 May 2012. Copulations occurred among all seven recommended breeding dyads (Fig. 1 and Table 5). The average duration for copulation was 102 minutes (SD=64) and all copulations were completed (i.e. the pair had separated naturally) before staff left for the day. The majority of females that copulated did so during their first introduction; one copulated on the second attempt. Due to additional excellent breeding success at Mission Creek, the Oregon Zoo did not conduct breeding introductions with the back-up breeders. In fact, on 2 May 2012, all back up breeders were transferred to Mission Creek for inclusion in their captive breeding. In addition, 12 surplus butterflies from four OZ primary breeder matrilines (2 dyads) were transferred to Mission Creek. These animals were not needed for the captive colony and their transfer supported increased sample sizes for the oviposition study (See Expand captive propagation at Mission Creek).

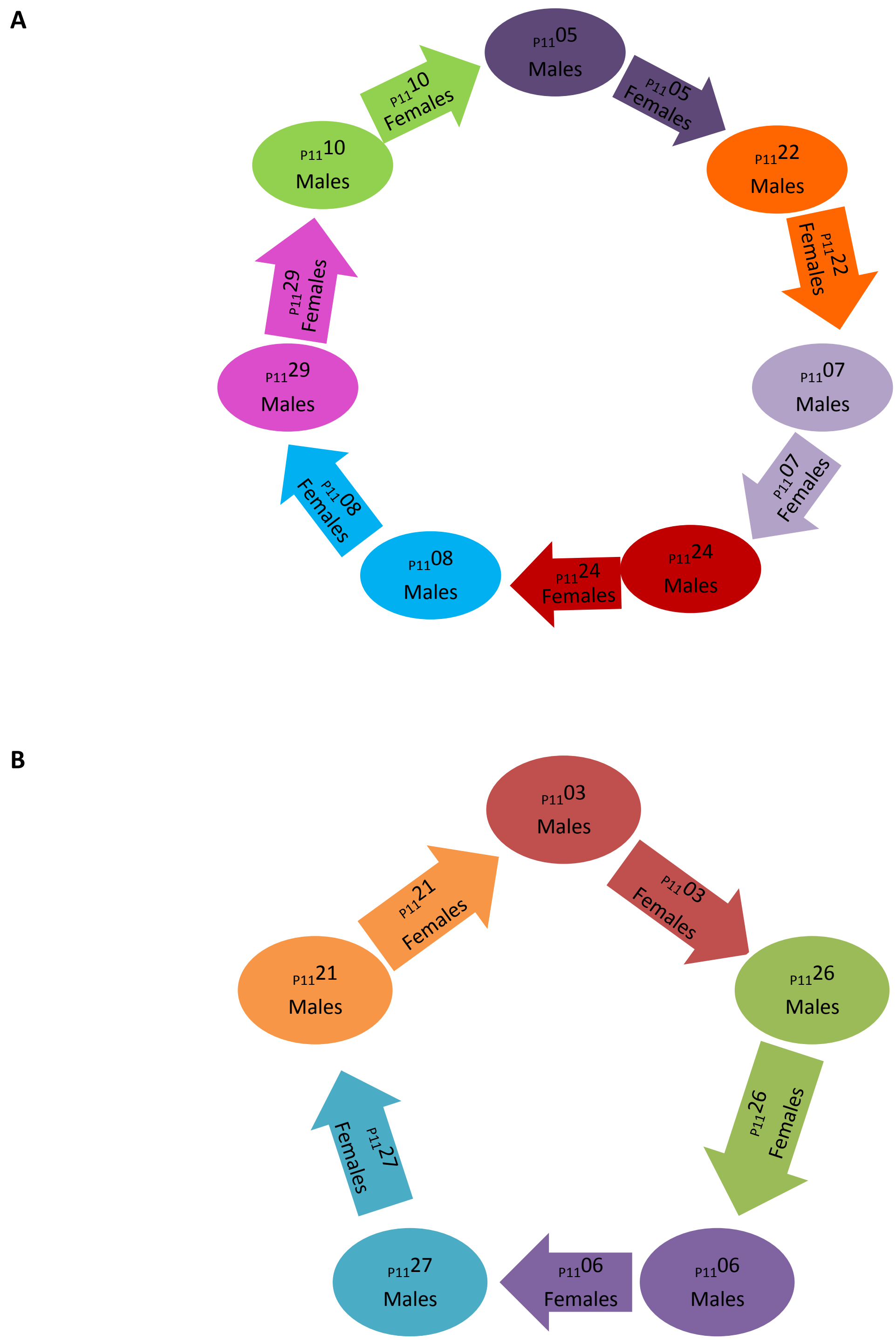


Fig. 1. “Daisy chain” pairing strategy used to manage breeding introductions in 2012 at A) the Oregon Zoo, Portland, OR and B) Mission Creek, Belfair, WA, within the captive colony co-managed by these institutions (see Lewis et al. 2013, *in prep*.).

Captive rearing egg to adult

Most captive females laid eggs in 2012 (Table 5), however many wild females failed to lay (Table 6). In addition, a total of 11 gravid females from the Oregon Zoo and Mission Creek that were sent to the field for release (Table 7), were returned to the Zoo to continue ovipositing when it was learned that egg production of wild females remained critically low (Table 6). A total of 8,891 eggs were produced between the two facilities with 69.4 percent (6,173 eggs) produced from known lineage captive-mated females, 7.4 percent (656 eggs) produced from wild females, and 23.2 percent (2,062 eggs) produced from captive females of unknown lineage. Two females (one captive-mated and one wild) at the Oregon Zoo laid a total of 298 eggs that did not hatch; these data are currently unavailable for Mission Creek. These results exceed our target production of eggs for 2012 (5,600 total), although the contribution from wild females was well below our expectations (3,735 or two-thirds of the total). At the Oregon Zoo, wild females laid less than half as many eggs per female as captive-mated females (Tables 5 and 6). Similar data are not currently available for Mission Creek, but judging from the number of eggs produced by wild females (Table 6) the same trend held there.

Table 5. Captive-mated Taylor's checkerspot females/matrilines, copulation date and time, number of oviposition days, and oviposition outcomes to diapause at the Oregon Zoo, Portland, OR, and Mission Creek Corrections Center for Women, Belfair, WA, 2012. Number hatched is taken at 3rd instar due to the small size and tight clustering of early instar larvae (from Lewis et al. 2013). Summary statistics (average and standard deviation) are provided for females with eggs that hatched at the Oregon Zoo.

Female ID	Date	Time (min)	# days	#eggs	Avg. % developed	# hatched	# in diapause
Oregon Zoo – by individual							
11FL29-05	10-May	130	6	178	0	0	0
11FL07-04	12-May	85	2	169	100	141	139
11FL10-03	13-May	42	14	151	16	35	35
11FL29-14	14-May	55	14	99	8	2	1
11FL24-04	15-May	80	1	228	100	154	115
11FL08-05	15-May	100	2	130	100	89	88
11FL24-05	16-May	105	1	154	100	144	142
11FL22-12	16-May	135	1	145	100	112	112
11FL22-19	19-May	35	18	180	48	55	53
11FL07-19	19-May	100	2	187	100	152	145
11FL10-22	19-May	105	2	157	100	147	147
11FL08-12	19-May	115	8	433	100	427	419
11FL05-08	19-May	135	12	99	75	50	47
11FL05-09	19-May	280	13	314	100	278	275
Subtotal	-	-	**90**	**2446**	**-**	**1786**	**1718**
Ave (SD)	-	**106 (61)**	**7 (6.4)**	**188 (92)**	**81 (34)**	**137 (112)**	**132 (110)**

Mission Creek – by matriline							
11FL03	-	-	-	502	-	368	356
11FL05[1]	-	-	-	354	-	408	407
11FL06	-	-	-	271	-	286	251
11FL21	-	-	-	302	-	302	300
11FL24[1]	-	-	-	434	-	434	432
11FL26	-	-	-	677	-	678	678
11FL27	-	-	-	1187	-	1137	1130
Subtotal				**3727**		**3613**	**3566**

[1] These lines brought from the Oregon Zoo for oviposition.

Eggs from wild and captive-mated females finished hatching in unison; those from females returned to the Zoo hatched last. Eggs from captive-bred females at the Oregon Zoo began hatching on 22 May and finished 12 June 2012; those from wild females began hatching on 1 June and completed hatching on 12 June 2012; those from returned females began hatching on 5 June and finished on 20 June 2012.

Table 6. Individual identification, number of oviposition days, and oviposition outcomes to diapause for wild female Taylor's checkerspot at the Oregon Zoo, Portland, OR and oviposition outcomes to diapause for all wild females at Mission Creek Corrections Center for Women in Belfair, WA in 2012. Eggs can be difficult to count due to stacking and should be viewed as a minimum estimate. Number hatched is taken at 3rd instar due to the small size and tight clustering of early instar larvae (from Lewis et al. 2013). Summary statistics (average and standard deviation) are provided for productive females at the Oregon Zoo.

Female ID	# days	# eggs	Avg. % developed	# hatched	# Into diapause
Oregon Zoo					
$_{P12}$FL01[1]	5	0	---	---	---
$_{P12}$FL05[1]	2	0	---	---	---
$_{P12}$FL06[1]	4	0	---	---	---
$_{P12}$FL08[1]	4	0	---	---	---
$_{P12}$FL11[1]	9	0	---	---	---
$_{P12}$FL02	10	77	100	40	40
$_{P12}$FL03	8	58	100	51	51
$_{P12}$FL04	13	91	84	70	69
$_{P12}$FL07	8	31	67	22	22
$_{P12}$FL09	11	70	83	74	71
$_{P12}$FL10	13	134	83	135	133
Total	**63**	**461**	**---**	**392**	**386**
Ave (SD)	**11 (2)**	**77 (35)**	**86 (12)**	**65 (39)**	**64 (38)**
Mission Creek					
Total		**195**		**229**	**227**

Numbers of prediapause larvae produced by female (Oregon Zoo) or matriline (Mission Creek) from captive-mated, wild and returned females are presented in Tables 5, 6 and 7, respectively. The greatest losses are typically observed from egg to hatching (defined as 3rd instar; Tables 5, 6 and 7), however after hatching, numbers changed very little. The first larvae at the Oregon Zoo entered diapause on 26 June and the last on 14 July 2012. In all, 6,543 diapausing larvae (5,932 captive and 611 wild) from the 2012 cohort were moved outdoors on 4 September 2012 to over-winter at the Oregon Zoo. In addition, 93 larvae from the 2011 cohort entered 2nd diapause in 2012 and were also placed in cold diapause.

A total of 6,589 postdiapause larvae (3330 from Mission Creek and 3,259 from Oregon Zoo) were provided to WDFW for release in 2013 (see Postdiapause release below); a few were found dead on release. In addition, 325 larvae from the 2012 cohort and 75 larvae from the 2011 cohort were retained in captivity to be included in the 2013 breeding colony (Table 8). Thus we exceeded our combined target production of 4300 postdiapause larvae for both rearing facilities by 62 percent.

Table 7. Individual identification, number of oviposition days, and oviposition outcomes to diapause for female Taylor's checkerspots returned to the Oregon Zoo, Portland, OR for continued oviposition in 2012 (see text for details). Number hatched is taken at 3rd instar due to the small size and tight clustering of early instar larvae (from Lewis et al. 2013). Summary statistics (average and standard deviation) are provided for productive females.

Female ID	# days	# eggs	Avg. % developed	# hatched	# Into diapause
$_{12}$RET-A	15	184	8	17	17
$_{12}$RET-B	12	243	80	166	163
$_{12}$RET-C	7	139	50	65	65
$_{12}$RET-D	9	201	100	149	147
$_{12}$RET-E	16	194	4	1	1
$_{12}$RET-F	17	223	100	216	215
$_{12}$RET-06A	18	120	0	0	0
$_{12}$RET-06B	18	339	69	184	176
$_{12}$RET-21-37	18	115	13	3	2
$_{12}$RET-26A	14	351	92	233	230
$_{12}$RET-26B	4	73	100	69	64
Total	**148**	**2,062**	**---**	**1,103**	**1,080**
Ave (SD)	**13 (5)**	**206 (89)**	**62 (40)**	**110 (90)**	**108 (88)**

The first pupa was found at the Oregon Zoo on 3 April 2013, 20 days earlier than in 2012, with the last pupa found on 2 May 2013. Postdiapause larvae averting the adult stage were showing signs of returning to diapause by 31 March 2013, a full month earlier than in 2012. In total, 40 larvae from 12 maternal lines entered 2nd diapause in 2013 (Table 8). This was less than half of the number in 2012; even so, some variation can be seen between matrilines.

A total of 294 pupae developed from the remaining 360 postdiapause larvae, 291 of which eclosed as adults (Table 8). Twelve had wings that eclosed improperly, but were still fit for breeding. Temperature, humidity, sunlight and airflow at the time of eclosion influence the unfolding and hardening of wings in adults both in captivity and in the wild; this condition is often unrelated to genetics. The first adult at the Oregon Zoo eclosed on 23 April and the last on 13 May; similar data are not currently available for Mission Creek.

Rearing conditions

Average minimum, maximum and range of temperature and relative humidity readings recorded at the Oregon Zoo and Mission Creek in 2012-2013 are presented in Table 9. Overall, temperatures at Mission Creek were remarkably similar to those at the Oregon Zoo, with Mission Creek trending towards slightly cooler at night and slightly warmer during the day, except when the Oregon Zoo was rearing in their outdoor enclosure (Gazebo – see Table 9). The similarities in rearing conditions that have been achieved are notable given that the lab at the Oregon Zoo is in a concrete block building, and the Mission Creek lab is in a greenhouse.

Table 8. Postdiapause larval numbers and outcomes by matriline for 2011 (2nd diapause) and 2012 Taylor's checkerspot cohorts retained for captive-mating at the Oregon Zoo, Portland, OR and at Mission Creek Corrections Center for Women, Belfair, WA, Spring 2013. 2nd diapause larvae avert development to the adult stage. Improperly eclosed adults (IE) are a subset of adults.

Matriline	# larvae	2nd diapause	# pupae	# adults	IE	Adults released
Oregon Zoo						
$_{11}$FL03	7	2	5	5	0	3
$_{11}$FL05	14	1	4	4	0	3
$_{11}$FL07	4	0	0	---	---	---
$_{11}$FL08	12	0	0	---	---	---
$_{11}$FL21	3	1	1	1	0	1
$_{11}$FL22	4	0	0	---	---	---
$_{11}$FL24	7	0	0	---	---	---
$_{11}$FL26	6	0	1	1	0	0
$_{11}$FL27	9	1	1	1	0	0
$_{11}$FL29	9	0	0	---	---	---
Subtotal	**75**	**5**	**12**	**12**	**0**	**7**
$_{12}$FL03	30	6	24	23	3	15
$_{12}$FL07	22	11	10	10	3	5
$_{12}$FL10	30	2	27	27	2	23
$_{12}$FL13	30	0	29	28	1	26
$_{12}$FL20	30	5	25	25	2	20
$_{12}$FL23	8	0	8	7	0	5
Subtotal	**150**	**24**	**123**	**120**	**11**	**94**
Mission Creek						
$_{12}$FL02	30	4	25	25	0	0
$_{12}$FL04	30	1	29	29	1	0
$_{12}$FL09	30	5	22	22	0	0
$_{12}$FL12	30	0	30	30	0	0
$_{12}$FL18	30	1	29	29	0	0
$_{12}$FL22	25	0	24	24	0	0
Subtotal	**175**	**11**	**159**	**159**	**1**	**0**

Expand captive propagation at Mission Creek using methods established at the Oregon Zoo

Methods

The original plan for 2012-2013 was to expand Mission Creek's rearing experience to include egg and immature larval stages in 2012 and allow them to dabble with captive-mating in 2013, scaling up to full capacity over a two year period. Because they exceeded expectations with rearing the mature stages in spring 2012 (Linders 2012b), they began to incorporate captive-mating techniques that same year.

Table 9. Average minimum, average maximum and (range) of temperature (°F) and relative humidity (%RH) during Taylor's checkerspot rearing by life stage, time frame and location at the Oregon Zoo, Portland, OR, and Mission Creek Corrections Center for Women, Belfair, WA, 2012-2013. Min/max data read daily at 1100 – 1300 h.

Life stage	Date range	Location[2]	Avg. temp	Avg. min temp	Avg. max temp	Avg min RH	Avg. max RH
Oregon Zoo							
Adult Males	04 May - 24 May 2012	Mezz.	-	64 (59 - 68)	74 (67 - 99)	42 (39 - 55)	56 (44 - 69)
Adult Females	07 May - 06 June 2012	Fridge C	-	33 (33 - 39)	43 (38 - 49)	26 (24 - 30)	51 (31 - 61)
Oviposition	10 May - 12 June 2012	Mezz.	-	66 (65 - 73)	78 (70 - 99)	48 (39 - 55)	58 (44 - 62)
Cooler Females	18 June - 24 June 2012	Cooler	-	61 (49 - 67)	70 (68 - 73)	35 (30 - 44)	61 (51 - 69)
Egg & Prediapause	22 May - 26 July 2012	Mezz.	-	68 (65 - 78)	75 (74 - 83)	50 (40 - 59)	62 (49 - 99)
Warm Diapause	14 July - 3 Sept 2012	Mezz.	-	69 (66 - 72)	78 (74 - 88)	50 (33 - 57)	77 (63 - 99)
Cold Diapause	4 Sept 2012 - 15 Feb 2013	Larvae Land	-	42 (29 - 53)	61 (40 - 82)	77 (53 - 96)	96 (70 - 99)
Wake-up	20 Feb – 3 March 2013	Mezz.	57	51 (47 - 57)	62 (57 - 65)	57 (45 - 68)	76 (31 - 88)
Post-diapause	4 March – 3 May 2013	Gazebo	52	42 (32 - 58)	65 (44 - 89)	42 (12 - 90)	90 (63 - 100)
Pupation	3 Apr - 16 Apr 2013	Gazebo	75	48 (19 - 74)	95 (87 - 101)	57 (49 - 67)	87 (79 - 93)
	17 April - 14 May 2013	Lab	70	65 (59 - 71)	80 (71 - 80)	43 (34 - 59)	84 (45 - 99)
2nd Diapause	31 Mar - 3 May 2013	Gazebo	54	42 (31 - 71)	70 (52 - 89)	36 (12 - 74)	91 (63 - 100)
	3 May 2013 – ongoing	Mezz.	69	65 (54 - 73)	74 (64 - 84)	59 (46 - 71)	76 (60 - 88)
Mission Creek							
Adult Males	27 Mar - 24 April 2012	Main	-	45.9	77.8	39.4	83.7
Oviposition	11 May - 26 May 2012	Main	-	58.2	83.7	28.9	66.5
Eggs & Prediapause	26 April - 10 July 2012	Main	-	56.6	82.3	31.8	71.9
Warm Diapause[1]	10 July - 11 Sept 2012	Main, low	72.7			(55.0)	
Cold Diapause[1]	12 Sept - 28 Feb 2013	Small	48.0			(83.6)	

Post-diapause	28 Feb - 22 April 2013	Main	-	45.9	65.5	42.4	69.9
Pupation	2 April - 22 April 2013	Main	-	52.0	71.7	46.1	71.2

[1] Data taken from Hobo data loggers in 8 oz rearing cups, to mirror conditions for larvae in diapause; RH is overall average.
[2] Location descriptions: Mezz. – mezzanine, a large, open, unfinished indoor area with minimal climate controls adjoining the main butterfly lab. Fridge C – A dedicated checkerspot refrigerator installed on the mezzanine. Cooler – a 48 qt Coleman insulated camping cooler housed on the mezzanine. Larvae Land - Outdoor overwintering area located under an overhang of the Animal Management building. Gazebo - a 12' X 12' screened enclosure with a vinyl roof located in a sunny area outdoors. Lab - Temperature and humidity controlled laboratory with a single Southeast facing window. Animals are housed on multi-shelved, rolling racks. Main room - 16 x 10-ft room in greenhouse at Mission Creek. Small room - a secondary 8 x 10-ft room at Mission Creek.

Projected full capacity for Mission Creek in 2013 was 1500 postdiapause larvae, 300 of which would be reared to diapause at the Oregon Zoo in 2012.

Captive rearing and captive mating techniques followed methods developed at the Oregon Zoo (Barclay et al. 2009); data forms were shared between facilities to enable consistent data collection. Target rearing conditions were the same as those for the Oregon Zoo (see Table 4). Due to differences in design between the two rearing facilities, we anticipated that modifications to the facility and/or the rearing protocol might be required to meet target conditions. We used an adaptive management approach at Mission Creek to improve the likelihood of success. This approach acknowledges that captive rearing programs consist of four dynamic components: 1) the rearing protocol, 2) the facility, 3) the staff, and 4) horticultural support, each of which can vary between facilities and require independent modifications to achieve success. Staffs from WDFW, the Oregon Zoo and Mission Creek maintained a close working relationship in order to identify and implement necessary changes to the protocols and facilities in a timely manner.

Results and Discussion

The Mission Creek facility continues to perform above expectation in nearly every way. In spring 2012 they successfully bred all five matrilines in their care (Fig. 1). They produced 3,727 eggs and put 3,566 larvae into diapause from captive-bred stock (Table 5). Mission Creek also improved care and feeding of early instar larvae by developing "Plantago pesto", a mash of Plantago leaves that was easier for them eat. Production of eggs from wild females was very low (195 eggs from 13 females), which was likely due to several factors. Wild females brought into captivity in 2012 were notably thin (see Collection of wild females, above). Being new to care of adults and oviposition, Mission Creek had to learn and develop techniques for coaxing eggs from females in poor condition. While data are not currently available for individual wild females, we also know from results at the Oregon Zoo that many wild females failed to lay; anecdotally this was also said to be the case at Mission Creek. They may also have spent less time on this activity because they were engaged in an oviposition experiment to look at the relative preference of Taylor's checkerspot for *Castilleja hispida*, *C. levisecta* and *Plantago lanceolata* (D. Aubrey, pers. comm.).

Because testing of a suitable diapause location was still underway at Mission Creek, 3,341 larvae were transferred from there to the Oregon Zoo for overwintering. Mission Creek retained 450 larvae in order to test their own diapause location in the small room of the greenhouse. Survival of these larvae during diapause was 99.1 percent and the remaining 446 larvae were ultimately released. A new diapause structure was also built at Mission Creek in 2013, which consists of a stand-alone wooden shed where environmental conditions can be kept cooler (Fig. 2).

In February 2013, 3106 postdiapause larvae were returned to Mission Creek for care until release; a subset was held to rear to the adult stage for breeding. Mission Creek maintained an excellent record while caring for large numbers of larvae, and at the close of the first complete year of rearing and breeding have demonstrated their ability to operate at full capacity with results comparable to the Oregon Zoo (see also Survival, below).

A few challenges do remain at Mission Creek, however. One facility-related challenge is heat build-up in the greenhouse, which required inmates to wear cold packs on their shoulders during the unseasonably warm weather we experienced in May 2013. Originally the greenhouse was designed with sliding windows and screens in the walls to be able to match ambient conditions as closely as possible when desired. Unfortunately, the DOC engineer that reviewed the plans felt the exhaust fans and roof vents would be adequate and this modification was unnecessary. We will continue to seek new solutions to address the issue of heat build-up. A second challenge has been getting the inmates access to a computer for data entry purposes, which would improve data sharing and summary. Fortunately it appears that a new computer will be in place at Mission Creek by year's end, and we anticipate that inmates will begin data entry at that time.

Fig. 2. New shed at Mission Creek Corrections Center for Women to be used for holding Taylor's checkerspot larvae through diapause, Belfair, WA, 2013.

Assess the efficacy of the captive propagation program and identify opportunities for improvement

Methods

Survival of captive animals

We used stage-specific survival rates to track success within and between years and identify areas for improvement. We calculated Kaplan-Meier (1958) survival rates for all captive stock between the following stages: egg, hatching (2nd instar), diapause, postdiapause, pupa, and adult; we also calculated the rate at which postdiapause larvae returned to diapause, averting development to the adult stage.

Adult measurements

Pupae and adults from each female line were measured and weighed using standardized procedures (Barclay et al. 2009) for comparison with measures obtained from wild adults. This is in keeping with best management practices (Crone et al. 2007) and allows us to determine if adults produced in captivity are undersized relative to their wild counterparts, which could result in reduced mobility in the field. Left side ventral hind wing area was calculated using ImageJ ver. 1.46r (Rasband 2012). Wing measurements will be compared between facilities and with those from wild adults using an ANOVA spreadsheet constructed by Shannon Knapp (WDFW biometrician) in Excel 2007. Left side ventral wing photos and hind wing area were also calculated for wild females brought into the lab for oviposition in 2012. Weights for wild females were not collected.

Results and Discussion

Survival of captive animals

As is typically the case, most mortality at both rearing institutions occurred prior to 3rd instar; this is also the stage at which survival varies the most from year to year. Both eggs and early instar larvae are difficult and time-consuming to count. Because we can expect survival to vary during this stage both in captivity (Linders, unpub. data) and in the wild (Kuussaari et al. 2004) a new simplified approach may be in order, whereby we get a reasonable estimate of the number of eggs, but avoid counting larvae until they enter diapause. During all remaining life stages survival remained in the 90th percentiles at both facilities (Table 10). Mission Creek, which began rearing checkerspots at the postdiapause stage in 2012, was highly successful in rearing the immature stages of Taylor's checkerspot. They also repeated their earlier success at rearing the mature stages in spring 2013. The high survival observed at Mission Creek from egg to hatch may be attributable to the "Plantago pesto" developed by the inmates for very young larvae; this technique may be too labor intensive to implement at the Oregon Zoo, but it is worth consideration. All survival rates were comparable between institutions and are well within the range of those reported previously (Fig. 3). Larvae returned to diapause at Mission Creek at a slightly lower rate than at the Oregon Zoo, although neither rate is very high. The high degree of success enjoyed across captive rearing institutions and years is very rewarding. Increasing survival of 2nd and 3rd diapause larvae was a goal in last year's report, which appears to be manifesting based on the results reported herein.

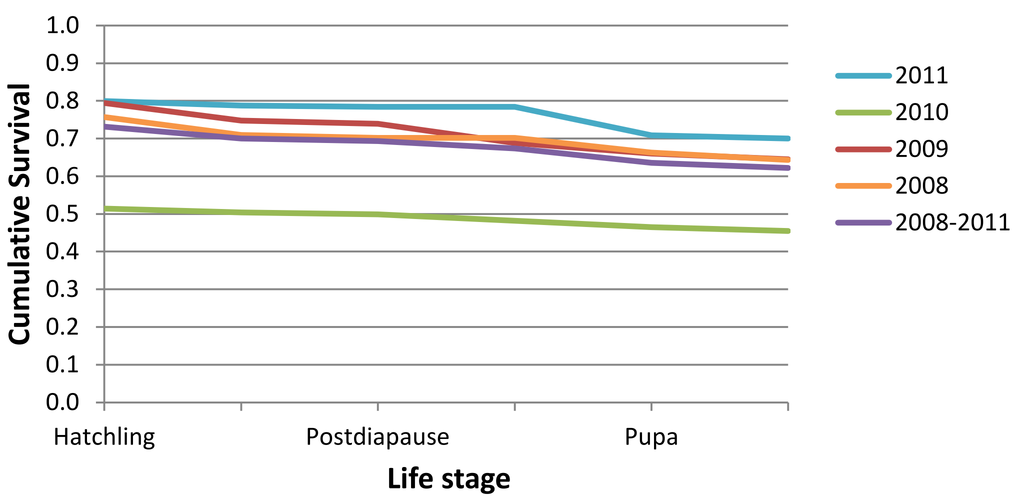


Fig. 3. Hatchling to adult survival for Taylor's checkerspots reared at the Oregon Zoo, Portland, OR, 2008-2011.

Table 10. Number of individuals and Kaplan-Meier (1958) survival by life stage for captive Taylor's checkerspot butterflies at the Oregon Zoo (OZ), Portland, OR and Mission Creek Corrections Center for Women, Belfair, WA, 2012-2013. Only clusters with some eggs that hatched are included here; number of hatched larvae was recorded at 3rd instar. Label abbreviations are 2011 (11) and 2012 (12) cohorts from Joint Base Lewis-McChord (FL); and RET = captive females that were brought back to OZ rather than being released.

Life stage or *Disposition*	2nd diapause 11FL		OZ Wild Caught 12FL		MC Wild Caught 12FL		Captive-bred 12OZ		Captive-bred 12RET		Captive-bred 12MC		All 2012 stock	
	#	% Stage survival	#	% Stage survival	#	% Stage survival	#	% Stage survival	#	% Stage survival	#	% Stage survival	#	% Stage survival
Eggs			461	---	195	---	2446	---	2,062	---	3727	---	8891	---
Eggs to hatch			392	85.0	226	---	1786	73.0	1103	53.5	3603	96.7	7110	80.0
Hatch to warm diapause	93	---	386	98.5	226	100.0	1770	99.1	1080	97.9	3554	98.6	7016	98.7
Warm to cold diapause at OZ	88	94.6	386		225		1753		1080		3099	99.5	6543	93.3
***Larvae to MC for wake-up*[1]**	---	---	*228*		*141*		*698*		*541*		*1944*		*3552*	---
MC Wake-up to release[2]			127	92.0	53	94.6	688	99.0	533	98.5	*1925*	99.0	3330	98.6
Larvae @ OZ for wake-up			158		84		1055		539		1601		3437	---
OZ Wake-up to release[2]	20	22.7	*76*	*100.0*	*16*	*100.0*	1031	98.5	538	99.4	1598	99.9	3259	99.1
***OZ/MC Captive breeding colony*[3]**	20		82/90	---	68/85	---								
Return to diapause	5	25.0	24/11		16.0/6.3									
Pupae	12	60.0	123/159		97.6/97.0									
Adults	12	100.0	120/159		97.6/100.0									

[1] Includes 450 larvae diapaused at MC, 446 of which (99.1) survived.
[2] Two week timeframe.
[3] OZ Wild and MC Wild combined by institution from here; # is the column on the left for OZ/MC, and % stage survival is the column on the right for OZ/MC.

Adult measurements

Not all data needed for the ANOVA are currently in hand; analyses comparing adult and pupal size between institutions and the wild will be run once all data are in hand. Weight and wing area measures for male and female checkerspots reared at the Oregon Zoo and females brought in from the wild for oviposition are presented in Table 11. Adult measurement data for captive adults from Mission Creek in 2012 is forthcoming.

Table 11. Average, range and number of captive-reared Taylor's checkerspots measured at the Oregon Zoo, Portland, Oregon, with regard to pupal weight, adult weight, left ventral wing area. Left ventral wing areas are also presented for wild females brought to the Zoo for oviposition in 2012.

	Average	Range (g)	n
Female pupal weight	0.220 g	0.164 – 0.280	77
Male pupal weight	0.175 g	0.110 - 0.221	67
Female adult weight	0.150 g	0.114 – 0.206	77
Male adult weight	0.087 g	0.051 – 0.129	67
Female left ventral wing area	1.669 cm^2	1.137 – 2.485	66
Male left ventral wing area	1.249 cm^2	0.806 – 1.848	61
Female left ventral wing area (wild collected)	1.853 cm^2	1.359 – 2.300	16

III. Release captive and associated wild stock

Postdiapause larval release 2013

Postdiapause larvae are the preferred stage for release because they are robust and nearly mature. Larvae were brought to the field packed in labeled deli containers containing freshly-cut leaves of *Plantago*; containers were packed in coolers without ice or heat. A total of 3,259 postdiapause larvae were released at Scatter Creek South on 1 March 2013. On 4 March 2013, a total of 3,330 larvae were released at Glacial Heritage Preserve. Four hundred twenty of the GHP larvae were included in a release experiment involving two burned and two unburned plots, one in each treatment containing *Plantago lanceolata* and the other *Castilleja levisecta*; larvae were equally distributed between treatments. Results of this trial will be included in a manuscript currently in prep. Larvae at all locations were released in groups of 2-5 on large and/or dense host plants/patches (*Plantago lanceolata, Castilleja hispida* or *Castilleja levisecta*) within restored prairie. Weather on both release days was excellent, with temperatures ranging from 5.0-14.0 °C, average wind speed ranging from 1.5-8.0 mph, clear to cloudy skies and distinct to no shadow; cloudy conditions corresponded with warm temperatures. At least 10 people assisted with releases at each site, working from about 1000-1630 to complete the task.

Adult release 2013

A total of 107 adult Taylor's checkerspots not needed in mating trials and females that had achieved oviposition targets in captivity (i.e., sufficient to represent their lineage) were released at GHP (Appendix A) on 19 May 2013. As in previous years, adults were transported in net enclosures and released directly into the environment by allowing them to fly from the cage or placing them on a nectar plant (Fig. 4). Weather at the time of release was 17.0 °C, wind speed averaged 1.1 mph, and skies were overcast and bright with no shadows.

IV. Monitor success of the reintroduction

Documenting presence and relative abundance through various life stages provides near-term measures of survival and improves the likelihood that factors affecting success will be detected. Over the longer term, population targets and population monitoring goals will be used to evaluate success in population establishment and demonstrate progress toward species recovery.

Document postdiapause larval presence and relative abundance in release areas

Past releases have shown that larvae and adults exhibited normal behaviors immediately following release (e.g., feeding, basking, mating, and ovipositing). We have also relocated animals in release areas in the days, weeks, months and years following release, giving us high confidence that animals are surviving and in most cases, reproducing.

Fig. 4. Captive-reared, unmated male and female Taylor's checkerspots co-housed prior to release to stimulate mating (left); copulating pairs are visible in the enclosure. At right a checkerspot being hand-released in South Puget Sound, Washington, May 2012.

Methods

To confirm site occupancy, quantify relative abundance of larvae, and identify issues that may be cause for concern, we conducted surveys for postdiapause larvae in active release areas on two occasions in the weeks following release (weather permitting). All larvae were counted within randomly selected 5 x 2-m belt transects, spaced 5-10 m apart on an east-west orientation through release areas to neutralize visibility. Surveys from 2009-2012 were conducted in square or rectangular plots with standardized transects occurring at 5-m intervals. Release plots in 2013 were amorphous, which required us to adjust the manner in which transect start locations were selected. For example, we would approach a plot from the southeast edge and continue walking north until we encountered host plant, indicating we were within the habitat. At that point we began surveying in 2 x 5-m segments heading west until we reached the edge of the plot or ran out of habitat. Surveys were conducted on days with temperatures >8 °C, wind <15 mph, and at least a soft shadow; narrowing these variables further risks finding sufficient time to conduct surveys. Initial surveys were conducted not sooner than the day following release or as soon thereafter as possible, with a second survey conducted at least 1 day after the first survey or as soon thereafter as possible. A third survey was conducted in 2013 because weather conditions remained favorable and larvae remained active. This basic method has been used since 2009. Data for all sites from 2009-2013 are presented herein. All 2013 surveys results are reported, but only data for the most productive surveys (one per site per year) are presented for 2009-2012. This approach was also used in the regression analyses due to the influence of weather and larval behavior on detectability (see 1 x 1-m plot analysis below). Surveys were discontinued once most larvae had reached sixth instar (about 2.5-3.0 cm in length) because at that point they are likely to pupate or return to diapause, affecting their detectability. Our main objective in estimating abundance is to assess survival relative to the number of larvae released and evaluate relative success between reintroduction sites and years.

Data analyses. To calculate estimated abundance of larvae observed, counts for each 5 x 2-m sample-plot were expressed as densities (#/m^2) then averaged across all sample plots by site and year. Mean density was extrapolated to the total release area and standard errors calculated. Linear regression was performed in MS Excel 2007 using the most productive survey from each site/year.

1 x 1-m plots

To track numbers of larvae post-release in a more controlled setting, five postdiapause Taylor's checkerspot larvae were released in each of ten 1 x 1-m^2 plots at Scatter Creek South on 3 Mar 2010. On the day of release, one observer surveyed the plots three times: 30 minutes, 60 minutes, and 2 hours post-release. Plots were also revisited at two time intervals (1100 and 1300) on the day following release (4 Mar), and once around midday (1100-1500 h) on Days 3, 5, 8, 11 and 15 post-release (5, 7, 10, 13 and 17 Mar). Each plot was searched for 1 minute and the number of larvae recorded; when fewer than five larvae were observed, one additional minute was spent searching 0.5 m beyond the plot perimeter. Weather on survey days was above 8.0 °C with little or no precipitation.

Results and Discussion

Three complete surveys were conducted at GHP on 9, 18-19 and 25-26 March and at SCS on 8, 15-18 and 24&27 March 2013. Typically more than one day was required to survey an entire site due to the short duration of suitable weather in March and the expanded scope of the releases in 2013. All surveys met protocol conditions except small portions of the day on 15 and 19 March, when there was insufficient sun to cast a shadow. Winds were generally calm, averaging 6 mph or less, except for a portion of the day on 18 March at SCS (12 mph ave); temperatures ranged from 8-18 °C. Larvae were readily observed in most plots at both sites until they entered the pupal stage in early April.

Post-release survey results relative to release size and date are presented in Table 12. As expected, numbers detected were generally lower in the days following release, although 2013 surveys suggest better than average results, especially at GHP. The degree to which this results from detectability vs. mortality and dispersal, however, is unknown. Unfortunately, even surveys designed to measure differences in detectability between observers cannot easily parse the relative contribution of observer detectability vs. larval detectability caused by change in position (e.g., basking on top vs. on the underside of a leaf). Surveys for postdiapause larvae are challenging to implement due to the very short periods of suitable weather typical for western Washington in March. As result the "best" post-release surveys varied in timing from 5 to 26 days post-release (Table 12). Restricted access to R50 in 2011 resulted in a late release which occurred when larvae were already mature (20 March) with post-release surveys conducted after some larvae had already pupated (16 April), making those numbers unreliable. Surveys in 2012 were conducted somewhat later due to an initial change in methodology, which was later abandoned. Somewhat unusual results were obtained in 2013, with the last surveys being the most productive. The abundance estimates for this time period may be somewhat inflated, as larvae appeared to congregate (note the increase in estimated density at GHP relative to the release density); in some cases more than 10 larvae were found within one square meter.

Table 12. Number (N) and density (per square meter) of postdiapause larvae released; number of sampling units, count, density, standard error, and estimated abundance (n) of larvae observed on post-release surveys by site and date. Site codes are: SCN = Scatter Creek North; SCS = Scatter Creek South; PCM = Pacemaker; GHP = Glacial Heritage Preserve; R50 = Range 50; South Puget Sound, Washington, 2009-2013.

Site	Release date	N	Release density	Survey date	# units	Count	Density /m sq	SE	n
SCN	6-Mar-09	759	0.84	13-Mar-09	10	32	0.10	0.06	90
SCS	6-Mar-09	747	0.83	12-Mar-09	10	17	0.06	0.02	51
SCS	3-Mar-10	891	0.45	8-Mar-10	8	40	0.09	0.04	187
SCS	6-Mar-11	1109	0.92	17-Mar-11	5	9	0.18	0.07	216
SCS[1]	1-Mar-13	3259	1.08	27-Mar-13	21	35	0.17	0.08	503
SCS	1-Mar-13	3259	1.08	27-Mar-13	30	96	0.45	0.05	966
SCS	1-Mar-13	3259	1.08	27-Mar-13	28	173	0.60	0.11	1802
PCM	6-Mar-09	741	0.82	13-Mar-09	10	40	0.13	0.02	120
PCM	8-Mar-12	1565	0.70	23-Mar-12	29	65	0.22	0.05	504
R50	4-Mar-10	1145	0.58	18-Feb-10	7	83	0.29	0.09	573
GHP	7-Mar-12	975	0.65	24-Mar-12	20	36	0.18	0.05	270
GHP[2]	4-Mar-13	2910	1.23	9-Mar-13	31	215	0.69	0.12	1637
GHP[2]	4-Mar-13	2910	1.23	19-Mar-13	28	199	0.71	0.15	1677
GHP[2]	4-Mar-13	2910	1.23	26-Mar-13	26	327	1.26	0.20	2968

[1] No samples taken in Plot I on this date.

[2] Larvae released in Plot D and area of Plot D excluded; not sampled as part of this analysis.

As expected, a regression of estimated abundance vs. number of larvae released for all sites from 2009-2013 (Fig. 5) indicates a strong positive correlation (Regression: $\hat{y} = -664.3 + 0.95x$; $R^2 = 0.84$; $F = 42.16$; $p = 0.0002$). To assess the influence of the large releases and high survival experienced in 2013 on the regression equation, a second regression was run without these data. The relationship between the number of larvae released and estimated abundance remains strong when only smaller releases are considered (Regression: $\hat{y} = -314.4 + 0.57x$; $R^2 = 0.70$; $F = 14.16$; $p = 0.009$). Points above the line indicate better than average sites and/or years. Among sites with more than one release, no one site exhibits a strictly below average trend. Nor are releases within years clustered consistently relative to the line with the possible exception of 2009, suggesting that in most cases outcomes are influenced by variability in a combination of biotic (site) and abiotic (microsite, weather) conditions.

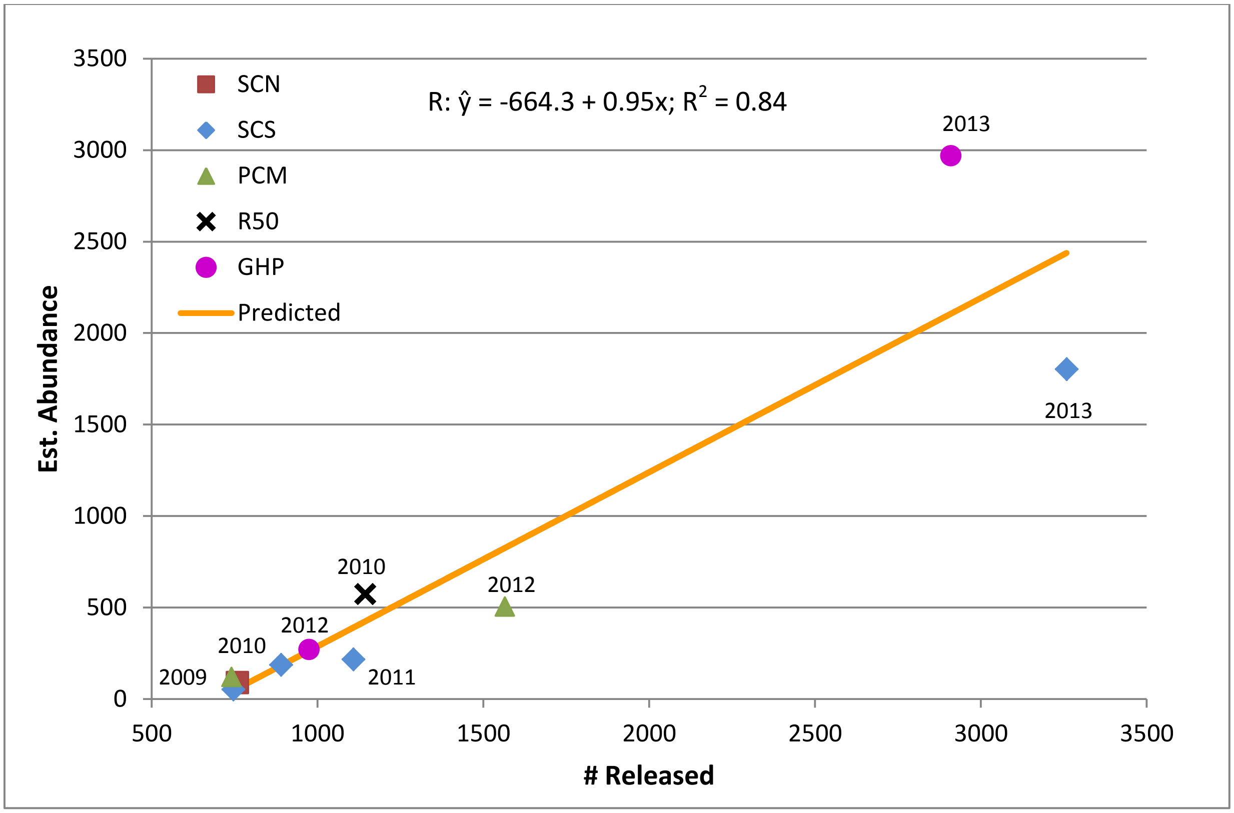


Fig. 5. Numbers of postdiapause Taylor's checkerspot larvae released vs. estimated abundance by site in South Puget Sound, Washington, 2009-2013; each symbol represents one site survey in one year.

1 x 1-m plots

All one meter square control plots were surveyed on eight days in 2010, for a total of 130 plot visits, or 13 surveys per plot. Of 207 total detections, 36 percent (75 larvae) were on the larval host, *Plantago lanceolata*. Almost 40 percent of larvae (81 of 207) occurred within 10 cm of a *P. lanceolata* plant; 18 percent (38) were >10 cm from *Plantago*. The number of larvae detected (Fig. 6) was negatively correlated with time post-release ($\hat{y} = 2.42 – 0.14x$; $R^2 = 0.62$; $F = 17.58$; $p = 0.002$). The most dramatic decline in detections occurred within the first 24 h post-release (Fig. 6), with only 25 of the 50 larvae (50%) detected 2 h post-release. Numbers were variable over the next several days, but on Day 5 returned to a level similar to that observed 2 h post-release. On occasion, larvae were observed basking on the undersides of leaves or curled on the ground surface beneath overhanging vegetation. Crypsis, or the ability of animals to avoid detection, is likely an anti-predatory adaptation contributing to low detection rates. A total of 14 larvae were observed beyond the plot perimeters. Distance from the plot edge ranged from 1 to 364 cm, with more than half detected within 50 cm of the plot edge. The larva detected 364 cm from the plot was observed on 7 March 2011, 5 days post-release, indicating that larvae can move considerable distances within a few days and that emigration also contributes to the decline in larval detections. Because crypsis and emigration reduce detectability of larvae, larval surveys can only be used to generate minimum estimates of abundance, which should not be equated with larval survival. These data suggest that multiple post-release surveys have two primary benefits: to document persistence over time and to establish greater confidence in minimum estimates of abundance that may be affected by details of the sampling protocol that are difficult to control (e.g., larval activity, interaction of weather variables, etc.).

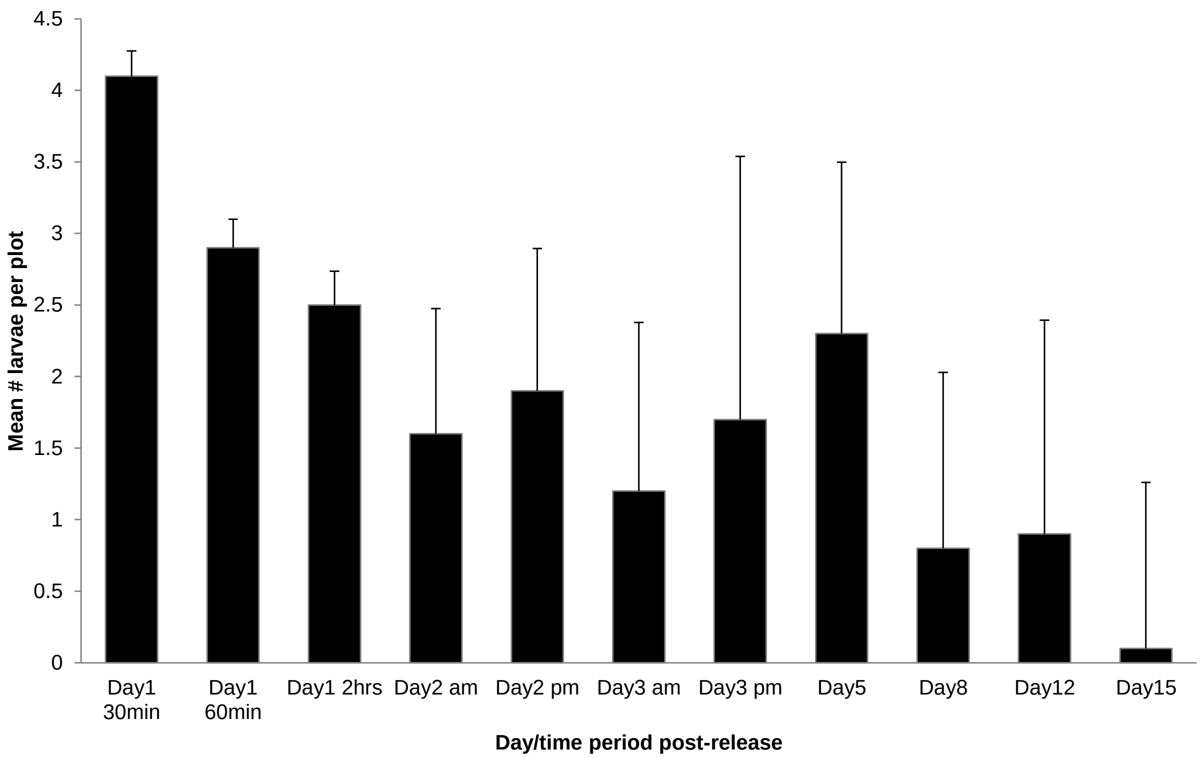


Fig. 6. Mean number (and SD) of Taylor's checkerspot larvae detected in ten 1 x 1-m plots by day and time period since initial release on 3 March 2010 at Scatter Creek South, in Thurston County, Washington.

Evaluate reintroduction success based on presence, relative abundance and distribution of adults

Methods

Adult presence and relative abundance

We used line transect sampling to quantify daily density, daily population size, and to illustrate the distribution of adults within the sampling area annually. Four reintroduction sites (SCS, R50, PCM and GHP) plus one extant site (R76) were surveyed for adult checkerspots during the 2013 flight season. Field sampling methods followed Linders and Olson (2012). Distance sampling was conducted up to 3 times per week during the flight season with a target minimum of 5 good sampling days per site. More surveys are preferable, including surveys with no detections to bracket the beginning and end of the flight season. Survey transects at reintroduction sites included all release plots and a buffer of sufficient size (200 m) to capture the anticipated adult use area, except at R50, where access to some areas is restricted. Transects at R76 covered the majority of the occupied area to which we have access. Transect length and spacing by site and year is shown in Table 13. The closer transect spacing at reintroduction sites insures that a sufficient number of butterflies are detected to calculate abundance estimates. Protocol surveys are those with ambient temperatures >11.7 °C (53.0 °F); sufficient sunshine to cast a soft (fuzzy) or distinct (sharp-edged) shadow OR bright skies with faint (can detect shadow but edges are nondescript) or no shadow if temp is >15.5 °C (60.0 °F); and sustained winds <10 mph. All surveyors received pre-season training and distance estimation skills were tested weekly throughout the flight season. Training ensures consistency in decision-making and survey technique and is critical for reducing the variance associated with density estimates.

We defined release success as the production of 10-30 adults per 1000 postdiapause larvae released based on the results of Harrison's (1989) work, but beyond the first year of release it is impossible to quantify how many adults originate from newly released larvae vs. those originating from reproduction in the wild. Instead, the presence and relative abundance of adults is an indication of year-to-year reintroduction success. Similarly, increases in the distribution of adults across the sampling area from year-to-year are an indication that the reintroduction is likely succeeding. It is not possible or perhaps even advisable to use an estimate of the annual population as a goal or a measure of release success both because we lack the methods to calculate such numbers and because the viability of a demographic unit may ultimately hinge on its ability to survive the population lows brought on by environmental extremes of drought and deluge (Ehrlich and Murphy 1987).

Data analyses. Analyses were conducted using Program Distance, Version 6.0 (Thomas et al. 2010) with density estimates computed for each survey date because population numbers can change daily. Detection functions were fitted using both the Conventional Distance Sampling (CDS) and the Multiple Covariate Distance Sampling (MDCS) engines. Summary statistics, including observation frequency tables calculated by observer and date, and sometimes by transect line, were calculated first. We also generated tables of encounter rates (observations per unit line length surveyed) by date and observer. For the MCDS analyses, we also computed univariate statistics and plots of distance data for potential covariates, such as observer, butterfly behavior, survey protocol, and weather, as recommended by Marques et al (2007). The best detection functions were chosen using a combination of default settings and user specified settings to select which of the many available models best fit the data, including Akaike's Information Criterion (AIC) and goodness-of-fit tests. More information on observer differences and detection functions used for 2009-2011 distance sampling analyses can be found in Linders (2010, 2011b, 2012b); data for 2012 are reported here. Data and density are presented for three reintroduction sites (SCS, R50, and GHP) plus R76 from 2012.

After determining the detection function(s) to use, density estimates were computed by date. Variance estimates of density were calculated using a relatively new method (Fewster et al. 2009) that takes advantage of the sequential (evenly spaced) layout of transects to reduce variance estimates over those, assuming that transects are placed randomly. Of the two methods of this type available in Program Distance, we chose to use method O2, which is generated by creating overlapping strata among adjacent transects and has been shown to increase precision with little change in bias (Fewster et al. 2009). Variances generated from the O2 method were used to estimate 95% Confidence Intervals. Density estimates were computed by survey date because of the expectation that population numbers change on nearly a daily basis due to eclosion and mortality of individuals.

Adult distribution

To illustrate distribution of adults within the survey grids at each site, all 2013 Taylor's checkerspot observations collected during distance sampling, regardless of date, were spatially joined to a GIS polyline layer representing transects and sections, then shaped into category classes and symbolized using a standard color ramp. Each category class was scaled so that the midpoint of each successive bin increased by a factor of two. Observations were overlaid on 2011 National Agriculture Imagery Program (NAIP) color aerial photos with 1 meter resolution.

Table 13. Number and length (m) of distance sampling transects and segments by site for extant (R76) and reintroduced populations of Taylor's checkerspot in the Puget Trough, Washington, 2010-2013.

Site	# transects	Transect spacing	# segments	Segment spacing	Transect length	Total line length	Survey area (ha)
2010							
SCS	11	25	11	50	600	6050	19.8
R50	13	25	8	50	400	5200	16.8
R76	12	50	14	50	700	8400	42.0
2011							
SCS	14	25	12	50	600	8400	22.5
R50	16	25	11	50	550[1]	8600	22.6
R76	12	50	14	50	700	8400	42.0
2012							
SCS	14	25	12	50	600	8400	22.5
R50	14	25	13	50	650[2]	8900	22.3
PCM	14	25	8	50	400	5600	14.0
GHP	12	25	8	50	400	4800	12.0
R76	12	50	14	50	700	8400	42.0
2013							
SCS[3]	14	25	7-12	50	350-600	5900	14.8
R50	16	25	13	50	650[2]	8900	22.3
PCM	14	25	8	50	400	5600	14.0
GHP	12	25	8	50	400	4800	12.0
R76	12	50	14	50	700	8400	42.0

[1] Two short transects measure 450 m each.
[2] Four short transects measure 450, 500, 550 and 550 m, respectively.
[3] Survey area was reduced in 2013 to focus on areas with highest sighting likelihood.

Results and Discussion

Adult abundance 2012

Scatter Creek South. Twelve surveys were conducted between 21 April and 3 June by four observers at SCS in 2012, but no butterflies were recorded prior to 7 or after 30 May (Linders 2012b); there were also no butterflies observed on 18 May prior to the release of adults (see Linders 2012b). In total, 87 butterflies were counted, with a peak daily count of 16, which was achieved on both 10 and 15 May (Table 14), despite over 98.4 km of transects surveyed in that time. Two of the four observers had similar levels of effort that were slightly higher than the remaining two. Allocation of effort per day was fairly evenly distributed, although the number of observers per day varied from 2 to 4. Regardless, the number of butterflies observed per survey date was too small to estimate detection functions by date, so a global detection function was fitted using CDS, and compared to observer covariate models in MCDS. There was very little difference in AIC between models. The CDS model had a slightly higher AIC, but was used for density estimation because it was simpler and estimates among all models were nearly identical. Density estimates with 95% Confidence Intervals are presented by date in Table 14 along with daily abundance estimates for the entire survey area.

Range 50. Nine surveys were conducted between 23 April and 3 June (Linders 2012b) by five observers in 2012 at R50; no butterflies were observed on the first and last survey dates (Linders 2012b). On all but 2 dates all transects were surveyed; on April 23 only 13 lines were surveyed (8250 m total) and on May 14 only 12 were surveyed (7,800 m total). While no observations were recorded on April 23, the reduced effort on May 14 must be considered when comparing density estimates, as the 2 lines not surveyed were edge lines that usually had fewer butterflies than did the other lines. In all, 2264 adults were counted, with a peak single day count of 709 (Table 14). Effort varied by observer with relatively even allocation of effort among observers, except one observer surveyed on only two dates; 3-4 observers surveyed on most days. Per-day sample sizes were large enough (Table 14) to fit date-specific detection functions. A series of CDS and MCDS models were analyzed, which looked at the effect of individuals vs. groups of surveyors and dates. The results indicated that detection functions differed by both observer and date. The best model was the CDS model by observer with detection functions by date, but date-specific estimates from this model are difficult to obtain. The best alternative to this was the MCDS model with date-specific detection function and an observer covariate. However, this model failed to produce reasonable variance estimates for April 29 and had overall larger variance estimates for the other dates. Finally, this model had a much larger proportion of variance in density due to the detection function, which has not been the case for other distance analyses of Taylor's checkerspot data. The next best model, a MCDS model with a global detection function and both date and observer covariates, had better variance estimates overall. It also had improved fit to the data for dates with fewer observations.

Pacemaker. Nine surveys were conducted at PCM between 28 April and 26 May (Linders 2012b) by four observers in 2012. A total of 91 butterflies were observed with a peak count of 21 on 10 and 13 May (Table 14); no butterflies were observed 28 April or 26 May. One observer had the greatest effort over the season with the other observers more similar to each other; all transects were surveyed on all dates. The number of butterflies observed per survey date was too small to estimate detection functions by date, so a global detection function was fit using CDS, and compared to observer covariate models in MCDS. The CDS model with a hazard key function and no adjustment terms fit best, and was used to compute daily density estimates.

Range 76. Eight complete surveys were conducted between 29 April and 29 May (Linders 2012b) by five observers at R76 in 2012. One observer surveyed only once, but most days there were 3-4 observers with effort fairly evenly distributed among them. In total 6,963 adult checkerspots were counted, with the greatest number of checkerspots (2,070) observed on 11 May. Three survey dates (29 April, 27 May, and 29 May) had <25 total observations each, so these dates were grouped to develop a single detection function. Per-day sample sizes were large enough (Table 14) to fit date-specific detection functions for all other dates. Attempts to include an observer covariate in that model failed. No other covariates were examined because preliminary summary statistics indicated no differences among behaviors, and there were no non-protocol observations in the 2012 data. The lowest total AIC among models for which daily estimates were computed was achieved by modeling the group of low observation dates with a global detection function and no covariates, and all other dates modeled independently with an observer covariate. The best detection function model for the group of low observation dates was one with a hazard rate key function and one cosine adjustment term. Detection functions for the other dates were all half-normal models with 1 or 2 cosine adjustment terms. Density estimates from the best models are shown in Table 14; models used by date are presented in Table 15.

Table 14. Raw counts, density estimates and adult abundance estimates including 95% Confidence Intervals for Taylor's checkerspot survey areas at Scatter Creek South (SCS), Range 50 (R50), Pacemaker (PCM) and Range 76 (R76) in South Puget Sound, Washington, Spring 2012. See text for details regarding derivation of density estimates.

		Density			Abundance		
Date	**Count**	**#/ha**	**Lower CI**	**Upper CI**	**#/survey area**	**Lower CI**	**Upper CI**
SCS							
7-May	7	0.81	0.30	2.16	18	7	49
10-May	16	2.42	1.01	5.78	54	23	130
13-May	6	0.80	0.26	2.53	18	6	57
15-May	16	2.10	1.01	4.37	47	23	98
18-May	0	0.00			0		
25-May	13	1.78	0.82	3.84	40	18	86
26-May	13	1.78	1.05	3.00	40	24	68
29-May	4	0.65	0.15	2.77	15	3	62
30-May	12	1.94	0.82	4.60	44	18	104
R50							
29-Apr	55	7.79	3.55	17.10	173	79	381
6-May	384	52.45	39.05	70.44	1168	869	1568
8-May	709	92.46	65.92	129.67	2058	1467	2886
11-May	494	66.08	52.42	83.31	1471	1167	1854
14-May	363	53.00	36.08	77.88	1034	803	1734
19-May	236	32.37	25.57	40.98	721	569	912
27-May	23	3.30	1.97	5.50	73	44	122
PCM							
2-May	1	0.18	0.02	1.31	3	0	18
7-May	18	3.19	1.69	6.03	45	24	84
10-May	21	3.19	1.39	7.35	45	19	103
13-May	21	3.37	1.04	10.97	47	15	154
15-May	14	2.59	1.30	5.19	36	18	73
18-May	10	1.60	0.58	4.37	22	8	61
19-May	6	1.06	0.36	3.18	15	5	45
R76							
29-Apr	20	5.37	2.21	13.08	226	93	549
6-May	1337	209.58	157.46	278.96	8802	6613	11716
8-May	1461	211.85	184.93	242.68	8898	7767	10193
11-May	2070	311.05	259.34	373.08	13064	10892	15669
14-May	1562	186.02	162.80	212.55	7813	6838	8927
19-May	480	63.72	51.26	79.20	2676	2153	3326
27-May	12	3.22	1.05	9.92	135	44	417
29-May	21	5.64	2.37	13.41	237	100	563

Table 15. Best detection function models by survey date for density estimation from distance sampling data collected at Range 76 in 2012. See Table 14 for associated density and abundance estimates.

Date	Model
29 April, 27 & 29 May	Global detection function over all 3 dates
6 & 8 May	MCDS with date-specific detection function and Observer covariate, w scale.
11, 14 & 19 May	MCDS with date-specific detection function and Observer covariate, σ scale.

Adult abundance 2013

Five to 8 complete distance sampling surveys were conducted at each site in 2013; four partial surveys each of R50, R76, PCM and GHP were also conducted (Table 16). Unlike in 2011 and 2012 (Linders 2011b, 2012b), flight season initiation and length appeared similar for all sites in 2013. Effort varied across sites, with the greatest effort expended at R50 (77,350 m) and R76 (56,000 m) where population size (Appendix B) and distribution (Appendix C, Fig. 2 and 5) are the greatest. Effort expended at three other reintroduction sites was lower (SCS: 50,150 m; PCM: 31,600 m; GHP: 40,000 m) but also reflects both population size (Appendix B) and distribution (Appendix C, Fig. 1, 3 and 4, respectively). The size of the survey area at SCS was reduced in 2013 to minimize the influence of areas with excessively low densities on population estimates; one complete survey of the 2012 grid was conducted on 14 May 2013 for distributional purposes, but no butterflies were observed outside of the standard 2012 survey area (Appendix B, Fig. 1).

Distance sampling surveys generated a combined total of 7,397 records across five sites (R76, R50, SCS, PCM and GHP) in 2013, with a total of 3,064 checkerspots counted (Table 16), comprising 2,896 groups. This is just one third of the number of checkerspots counted during the 2012 flight season (Linders 2012b). Butterflies were in groups of 1-3 with 2,737 single butterflies observed; 147 groups of two; and 11 groups of three. Distance estimates ranged from 0.0 to 18.0 m, with an average detection distance of 3.41 m across all sites (site-based averages ranged from 3.13 to 3.57 m), very similar to results in 2012. Checkerspots were present throughout the sampling areas at both R50 and R76 (Appendix C, Fig. 2 and 5), whereas distributions at SCS and GHP (Appendix C, Fig. 1 and 4) tracked the areas of release more closely. Adult counts were lower than expected based on postdiapause larval surveys (Table 12) and were also reduced relative to 2012 on sites where no releases occurred. Larvae continued to feed slightly later than they generally do and flight seasons timing was fairly typical. Consequently, the reduction in numbers across all sites would seem to be related to the warm temperatures and very dry conditions in South Puget Sound in early May 2013. Online weather data are now only available for a two month period, and were not currently available to assess conditions relative to other years. More work is needed to associate weather data and characteristics of flight season length, timing and stage-specific Taylor's checkerspot abundance, however monitoring of both postdiapause and adult stages is helpful in clarifying this relationship.

Table 16. Number of Taylor's checkerspots counted by site and date during distance sampling surveys at extant (R76= Range 76) and reintroduction sites (SCS = Scatter Creek South; R50 = Range 50, PCM = Pacemaker, GHP = Glacial Heritage Preserve) in South Puget Sound, Washington, Spring 2013.

Date	GHP	PCM	R50	R76	SCS	Comments
21-April			0			
22-April		0		0	0	
23-April	0			0		
26-April	21	0			21	
28-April			52	74		
01-May	57	0			64	
03-May			373			
06-May			208	865		R50: 11 of 16 lines; 5 lines-recorder lost
07-May	75				55	
08-May		0				
10-May			228	698		
11-May	60				33	
13-May		0				PCM: 9 of 14 lines surveyed, then rain
14-May			83		16	
16-May		0				
17-May	13					
19-May			10	39		R76: 9 of 12 lines
20-May	5				5	
25-May			8			
26-May	0					GHP: 4 of 12 lines surveyed; intermittent
01-June			1	0		
02-June	0				0	
Total	**231**	**0**	**963**	**1676**	**194**	**Number of observations**

Nectar observations were recorded opportunistically during distance sampling surveys for Taylor's checkerspot. Ten different nectar species were recorded across the four sites in 2013 (Table 17). The number of nectar observations in 2013 relative to 2011 and 2012 was notably reduced. This may be due to fewer observations of adults, but the drought that occurred in early May 2013 may also have been a contributing factor. As in previous years, three plant species, *Balsamorhiza deltoidea*, *Lomatium triternatum* and *Saxifrage integrifolia,* accounted for the majority (94.3 percent) of all nectaring observations in 2013; when both *Lomatium* species are included, 95.9 percent of all observed use is accounted for.

Table 17. Number and species of plant on which Taylor's checkerspots nectared during distance sampling surveys at three reintroduction sites (PCM = Pacemaker, R50 = Range 50, SCS = Scatter Creek South) and one extant site (R76 = Range 76) in South Puget Sound, Washington, 2012. ACMI – *Achillea millefolium*; ARMA – *Armeria maritima*; BADE - *Balsamorhiza deltoidea*; CAHI – *Castilleja hispida; CAQI -Camassia quamash*; CEAR – *Cerastium arvense* ; CYSC – *Cytisus scoparius;* ERLA - *Eriophylum lanatum*; FRVI - *Fragaria virginiana*; HYRA – *Hypochaeris radicata*; LECA – *Lepidium campestre*; LOSP – *Lomatium* species; LOTR- *Lomatium triternatum*; LOUT-*Lomatium utriculatum*; LULE – *Lupinus lepidus*; PLCO- *Plectritis congesta*; RAOC - *Ranunculus occidentalis*; SAIN - *Saxifraga integrifolia*.

Site	ACMI	ARMA	BADE	CAHI	CAQU	CEAR	CYSC	ERLA	FRVI	HYRA	LECA	LOSP	LOTR	LOUT	LULE	PLCO	POGR	RAOC	SAIN	Total
2011																				
R50	0	0	113																6	119
R76	3	14	618		3			12	46	1	1		621	5	3		3	4	37	1371
SCS	0	2	2		1								1			4				10
Subtotal	**3**	**16**	**733**	**0**	**4**	**0**	**0**	**12**	**46**	**1**	**1**	**0**	**622**	**5**	**3**	**4**	**3**	**4**	**43**	**1500**
2012																				
PCM			1									2	2					1		6
R50			86	1	4	1			3			13	4						30	142
SCS	1	2	6													1				10
R76		6	465		3		1		8			161	305	21				8	113	1091
Subtotal	**1**	**8**	**558**	**1**	**7**	**1**	**1**		**11**			**176**	**311**	**21**		**1**		**9**	**143**	**1249**
2013																				
GHP			2	1	5															8
R50			67							2			6					1	17	93
R76		1	219	1	1				2				48	6				1	3	282
SCS		1																		1
Subtotal	**0**	**2**	**288**	**2**	**6**	**0**	**0**	**0**	**2**	**2**	**0**	**0**	**54**	**6**	**0**	**0**	**0**	**2**	**20**	**384**
Total	**4**	**26**	**###**	**3**	**17**	**1**	**1**	**12**	**59**	**3**	**1**	**176**	**987**	**32**	**3**	**5**	**3**	**15**	**206**	**3133**
Percent	**0.1**	**0.8**	**50.4**	**0.1**	**0.5**	**0.0**	**0.0**	**0.4**	**1.9**	**0.1**	**0.0**	**5.6**	**31.5**	**1.0**	**0.1**	**0.2**	**0.1**	**0.5**	**6.6**	

Mardon skipper butterfly sightings were also recorded opportunistically during distance sampling surveys for Taylor's checkerspot at three of the sites visited in 2013 where they occur. In total 22 mardon skipper observations were made (Table 18), which is just 14.5 percent of the number detected in 2012. Most (14) were observed as singles; however, 4 groups of two individuals were also observed.

Adult distribution

Adults were distributed across 28.8 percent (34 of 118) of the segments surveyed at SCS (Appendix C, Fig. 1), with the greatest concentration of adults in the northwest quarter of the grid, where several of the most recent releases have occurred. Adults occupied 67.6 percent (134 of 198 segments) of the R50 survey area over the course of the flight season (Appendix C, Fig. 2). Although no sightings of Taylor's checkerspots were observed on distance sampling surveys at PCM in 2013, a few observations were noted in the area (Appendix C, Fig. 3). Adults were distributed across 48.0 percent (50 of 104) of the segments surveyed at GHP (Appendix C, Fig. 4), which is the greatest extent observed at this site since reintroductions began. At R76, the only extant site in South Puget Sound, 89.2 percent (150 of 168 segments) of the survey area was occupied during the peak single day count on 6 May 2012, with 94.0 percent occupancy when survey dates were combined (Appendix C, Fig. 5). While R76 surveys likely capture the core of the Taylor's checkerspot population at this site, the actual distribution is known to

Table 18. Number of mardon skipper butterflies observed by date and site during distance sampling surveys targeting Taylor's checkerspot butterflies in Spring 2013 at three sites in South Puget Sound, Washington. Only the first two "0" survey days for mardon skipper are included in the table. R50 = Range 50; R76 = Range 76; SCS = Scatter Creek South.

Date	R50	R76	SCS
6-May	0	0	
7-May			0
10-May	9	1	
11-May			2
13-May			
14-May	3		1
19-May	0	0	
20-May			1
25-May	5		
1-June	0	0	
2-June			0
Total	17	1	4

extend beyond the survey grid, as evidenced by observations at the survey area perimeter (Appendix B, Fig. 5; see also Linders 2012b). The same pattern is occurring at R50 (Appendix C, Fig. 2; see also Linders 2012b) to a lesser degree. However, as with the south edge of the R76 survey grid, access restrictions due to unexploded ordnance prevent us from expanding surveys in those directions. A single sighting at the north unit of Scatter Creek Wildlife Area (Appendix C, Fig. 6) and observations of checkerspots crossing the road to the south of the release area suggests dispersal from the site is likely occurring. Scattered sightings, including an early season observation of multiple individuals at R81 in the SE corner of the Artillery Impact Area (AIA), the 2,833-ha prairie that includes R76 and R50 (Appendix C, Fig. 7), suggests that the AIA may function as a large metapopulation if additional suitable habitat patches exist. Checkerspots were first observed in the vicinity of R81 in 2012.

Evaluate short-term success of releases based on reproduction

Methods

The first step toward establishing a naturally-occurring population is documenting local reproduction of released animals. We used 5 x 2-m belt transects placed randomly along transects at 5-10 m intervals to search for larval clusters in and around release plots on one newly established site (GHP). PCM was not re-surveyed due to the small number of adults observed there (Appendix C, Fig. 3) and because funding was sufficient to conduct surveys on only one site per year. Sampling units were oriented east-west to neutralize visibility; total line length was equal to about 10% of the survey area to standardize effort between sites. Surveys were conducted in late-June, when larvae were in about 2^{nd}-3^{rd} instar, on days with temperatures $\geq$10°C, average wind speed $\leq$10 mph, and at least faint shadows. Time, number of larvae in group, larval instar, host species, and distance from nearest oviposition site were recorded. Oviposition sites were typically revisited once every 7-10 days after they were located to document outcomes. Release plots were selected and enhanced to create high quality larval habitat, so we assumed these areas were also attractive to ovipositing females (Severns and Grosboll 2011). If no oviposition sites were located inside release plots, other suitable areas were searched if time allowed.

Similar surveys at R50 in 2010 located 29 oviposition sites in 534 square meters of search area, which is about 3 times the size of the area we searched; larvae at R50 were distributed across all release plots sampled. Based on results at R50 and the size of the area searched, we considered location of at least 10 larval clusters, where the majority of larvae and clusters persist to at least 3rd instar, a sign of successful reproduction.

Results and Discussion

Fifteen 5 x 2-m belt transects were searched across the six release plots at GHP between 21 and 27 June 2013. A total of 32 larval groups totaling 195 individuals were located. Groups ranged in size from 1 to 22 larvae ($mean$ = 6.1 + 6.0 SD; Fig. 7). Ten groups consisted of a single larva. Distribution of larvae by instar was: 14 first; 82 second; 26 third; 51 fourth and 22 fifth instar larvae. The fact that larvae were concentrated in second and fourth instars is thought to result from the release of adults near the end of the flight season. These results indicate the release at GHP exceeded our minimum threshold for success of 10 larval clusters. The proportion (37.4 percent) of larvae already at fourth instar or greater also indicates that survival of these clusters was likely high. In fact, at the end of the survey period there were so many mature larvae in the plots that it was nearly impossible to walk without risk of stepping on them and surveys were ultimately abandoned.

Fig. 7 Prediapause larvae resulting from reproduction of reintroduced Taylor's checkerspots at Glacial Heritage Preserve in South Puget Sound, Washington, Summer 2013. Larvae are in the fourth instar and are feeding on *Plantago lanceolata*, an exotic host.

Long-term monitoring and population goals

Methods

Abundance and distribution data from R76 are critical to understanding annual variations in population size and the shape and phenology of population curves (Weiss and Weiss 1998), which in turn help us set appropriate population targets, evaluate population growth potential, and set long-term monitoring goals. Monitoring will be used to measure progress toward population establishment 1) during the five-year release period, and 2) after the five-year release period is complete. For the purpose of this project, an established population is defined as follows:

> *A reintroduced population of Taylor's checkerspots will be considered established when a minimum of 250 adult butterflies (single day abundance estimate) are widely distributed across a monitoring area >20 ha (50 ac) in size and they occupy the site solely through natural reproduction each year for five consecutive years.*

The monitoring area at R76 covers about 42 ha (100 ac) or twice that of most reintroduction management units. The minimum single day abundance estimate is based on the peak single day abundance estimate at R76 in 2009 (Appendix C). In that year it was difficult to find 2 or 3 butterflies together on any given day, yet the population was able to recover from this point giving us confidence that other populations could as well. A five-year monitoring window is a widely used standard for establishing occupancy by butterflies. The monitoring scheme and distance sampling protocol already developed (Linders and Olson 2012) will be used as the basis for monitoring reintroduction sites.

Results and Discussion

Based on these criteria, R50 far exceeded the target of 250 adults on a single day on the first year in which the population was based solely on reproduction, with a peak single day count of 2,058 adults (range 1,467-2,886) in 2012. Raw counts for 2013 (Table 16) suggest abundance estimates will also exceed the target for 2013. In addition adults were distributed across the nearly the entire 22-ha monitoring area in 2012 (Linders 2012b) and 2013 (Appendix C, Fig. 2). Although they were rarely observed along the southern edge where checkerspot resources are lacking, they were observed at the northern edge of the survey area, where checkerspot resources extend beyond the survey grid.

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Future Plans and Recommendations

This is the eighth year of a multi-year recovery project. Translocation data from 2007-2013 are being compiled and preliminary results for many cross-year analyses are complete. Analyses on 2013 distance sampling data are scheduled for completion in winter 2013-2014. Plans for 2014 include continuing releases at Glacial Heritage Preserve (Thurston County) and initiating releases at Training Area 7S (JBLM). Given the robust response of the R50 population, no further reintroductions are recommended there at this time however population monitoring will continue in order to track the establishment and dynamics of the population over time. Funds for the 2013-2014 field season and rearing year have been secured; those for 2014-2015 will be requested from USFWS, ACUB and JBLM. Goals are to continue relatively large releases, while maintaining a number of release sites and microsites to offset the influence of climatic perturbations on translocation success. Additional sources of funding and new conservation partnerships are being explored.

While much progress is being made, more work is needed to achieve recovery of Taylor's checkerspot butterfly in Washington and throughout the species range. Following are several key issues that need to be addressed in the near term to improve our understanding of suitable habitat, functional population size, and dispersal capability of Taylor's checkerspot.

To increase the scientific basis of habitat enhancement planning and determination of site readiness for Taylor's checkerspot reintroductions, we need to quantify known habitat characteristics at occupied sites and at current and future reintroduction sites. To this end, the ACUB Taylor's checkerspot Habitat Characteristics project was initiated as a cooperative project with JBLM. That project got underway in Summer 2013 using a rapid assessment method to quantify resources at the site-scale using the 25 x 25-m grid format established by the ACUB Prairie Quality Monitoring project and the Land Condition mapping effort at JBLM, and is also used to collect distance data in this project. Data collection methods have been developed, data have been collected and results are being summarized, but additional funding will be required to expand the number of prairies and the size of areas included in the assessment. These data will be used to: 1) set quantifiable restoration targets, 2) measure progress toward achieving them, and 3) determine site readiness for reintroduction. This effort will insure the best use of limited restoration resources and increase the potential for reintroduction to succeed in an affordable and efficient manner.

Given many recent sightings of checkerspots in areas previously unknown, area search surveys are needed in the vicinity of reintroduction sites to document dispersal and potential colonization sites. In 2012 and 2013, checkerspots were sighted up to 2.5 km from known occupied sites on 13th Division Prairie and at Scatter Creek Wildlife Area. Documenting new breeding populations is especially important to recovery and will improve our understanding of checkerspot demography and ultimately reduce the expense of captive propagation and reintroduction as a path to increasing the size and number of populations. This work could be done by searching sighting locations and by reconnaissance of potential habitat in concentric circles outward from release areas. Any suitable habitat identified through this process could be mapped and periodically revisited.

Finally, additional funding will be required to develop a method for estimating total population abundance from the series of daily abundance estimates generated via distance sampling, as there are several exercises critical to our understanding of checkerspot demographics that cannot be answered using a series of daily abundance estimates. A key example of this is population modeling. An initial attempt was made to develop population projection matrices using existing data, however the lack of a single figure for the adult population made it impossible to understand the output of the model. Population estimates are also crucial to our understanding of how biotic and abiotic factors influence survival at different sites. This requires that restoration sites cover a broad range of climatic and microclimatic gradients, which will also maximize the potential for checkerspots to persist in the face of climatic perturbations and long-term climate change.

Questions for Further Research

1) What is the total population size of adult butterflies in any given year?
2) How does the distribution of adults relate to within-site habitat variables?

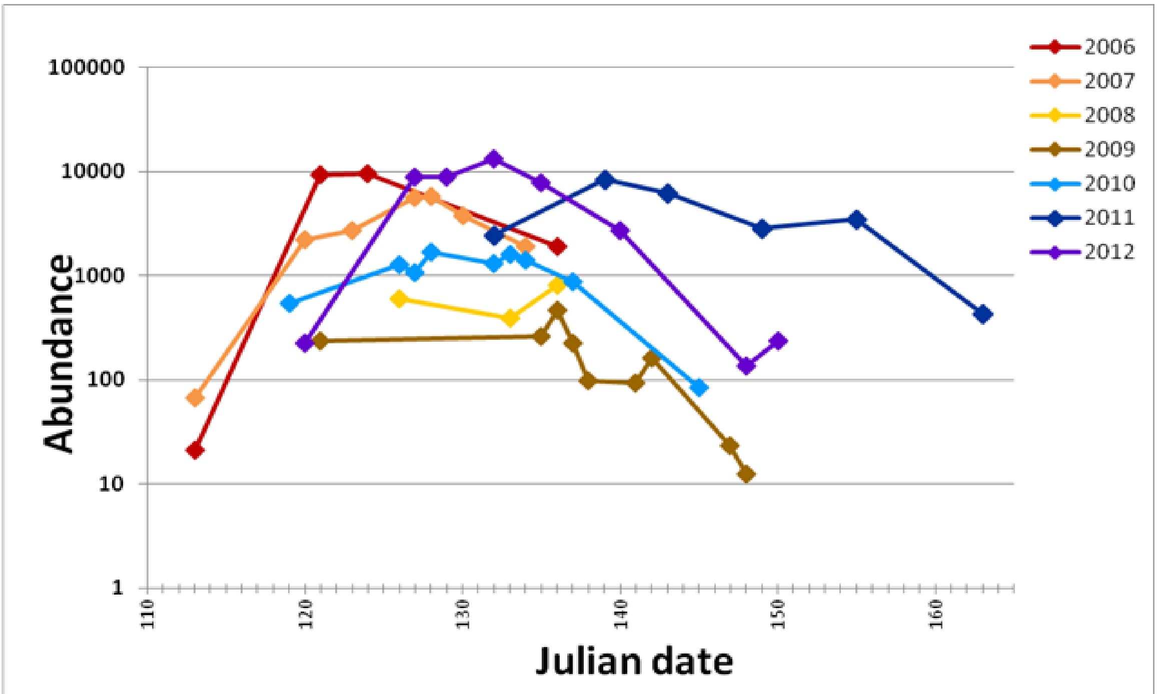
The first question has long been problematic because daily counts of butterflies, or abundance estimates based on such counts, reflect only a portion of the total population during any given flight season. Subsequent counts include some individuals that were part of previous counts, plus new individuals, such that numbers reflect emergence and death rates that vary through the flight season. Without estimates of these rates, estimating a total population size is difficult, but may be accomplished based on the pattern of the counts and some assumptions. Various methods have been used to address this problem, but these are often very sensitive to the validity of the assumptions.   Recent methods applying Bayesian modeling techniques to estimate salmon escapement appear to have application to the butterfly abundance estimation problem, and these are worth investigating as at least a starting point to developing a methodology for Taylor's checkerspot.

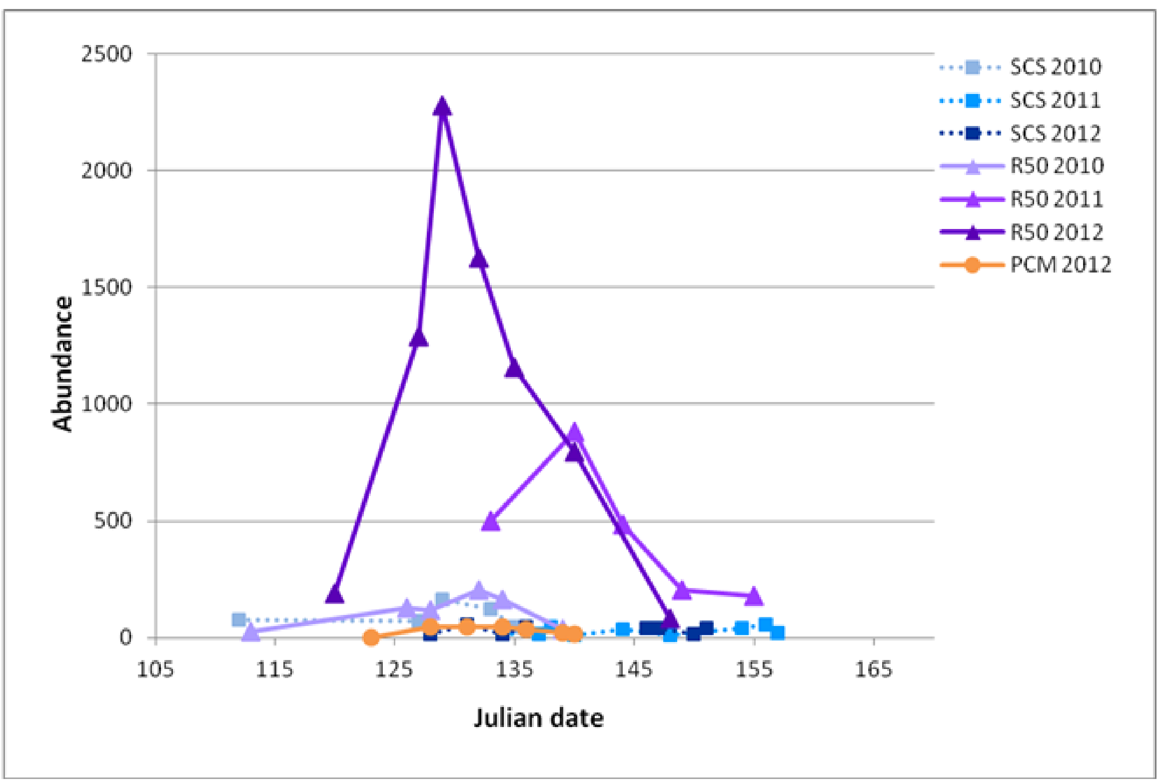
The second question may be addressed using existing Density Surface Modeling (DSM) techniques available in Program DISTANCE, the same software used to more generally estimate butterfly density from distance data.   DSM allows the modeling of the spatial pattern of detection-adjusted counts from distance sampling with respect to covariates collected over the same sample space. At Range 76 distance data are recorded by segment within systematically spaced transects, essentially forming a grid of count data. The availability of habitat data collected over this same grid, from RTLM or other habitat assessment protocols, enables the application of DSM to test hypotheses regarding the effects of habitat covariates on Taylor's checkerspot butterfly distribution.

Appendix A. Approximate number of Taylor's checkerspot butterflies released or proposed for release by life stage (PreD = prediapause larvae; PostD = postdiapause larvae) and site (SCS = Scatter Creek South; R50 = Range 50; GHP = Glacial Heritage Preserve; PCM = Pacemaker Airstrip; T7S = Training Area 7 South; SCN= Scatter Creek North; TNQ = Tenalquot Prairie; WPR – West Rocky Prairie; T15 = Training Area 15; BHP= Bald Hill Preserve).

	2006			2007	2008	2009		2010		2011			2012		2013		Proposed 2014	
Site	PostD	Egg	PreD	PostD	PostD	PostD	PreD	PostD	Adult	PostD	Adult	PreD	PostD	Adult	PostD	Adult	PostD	Adult
SCS		639		199	340	747	2487	891	202	1109	167	1036		133	3250			any
R50							2956	1145		1141			0		0			
GHP	30												975		3372	107	2000	
PCM			307			741							1565					
T7S			301														2000	
SCN						759												
TNQ																		
WRP																		
T15																		
BHP																		

Appendix B. Estimated daily abundance of adults based on distance estimation and timing of the Taylor's checkerspot flight season at R76 (Range 76 source site) in 2006-2012 (top) and at three reintroduction sites: SCS = Scatter Creek South and R50 = Range 50 in 2010-2012, and PCM = Pacemaker Airstrip in 2012 (bottom). The R76 survey area covered 42.0 ha in 2006-2012; note the log scale on the y-axis. R50 survey area was 16.8 ha in 2010, 22.6 ha in 2011 and 24.6 in 2012; SCS was 19.8 in 2010 and 22.5 in 2011& 2012; PCM was 12.0 ha in 2012.





Appendix C. Distribution maps of adult Taylor's checkerspots observed during distance sampling surveys at reintroduction sites (SCS, R50, PCM, and GHP) and one extant site (R76) in South Puget Sound follow. Each is labeled according to location, time frame and data displayed.

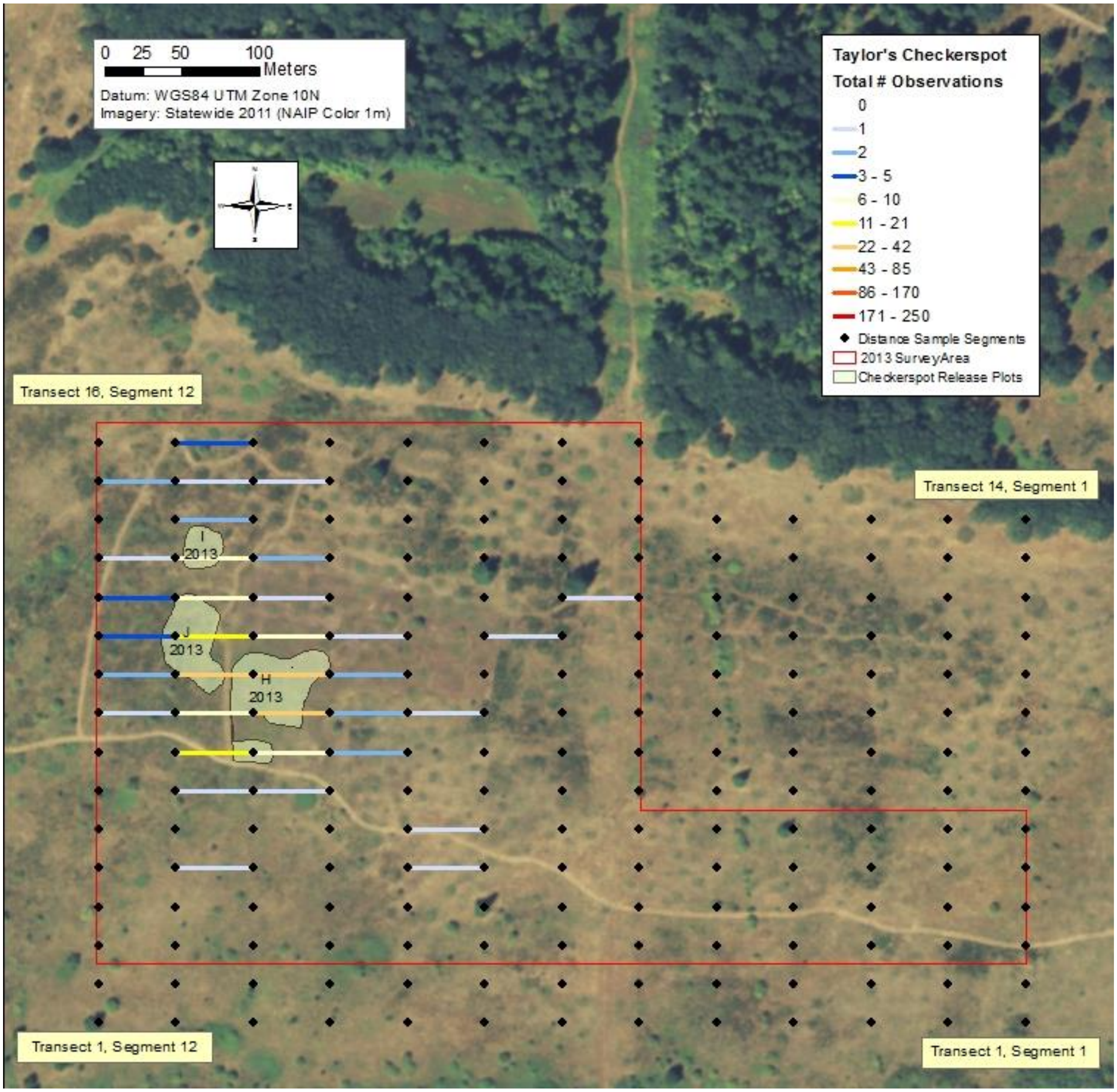


Fig. 1. Distribution of all adult Taylor's checkerspots observed during distance sampling surveys at Scatter Creek South, combined across all survey dates in Spring 2013, South Puget Sound, Washington. Red line indicated reduced area surveyed in 2013.

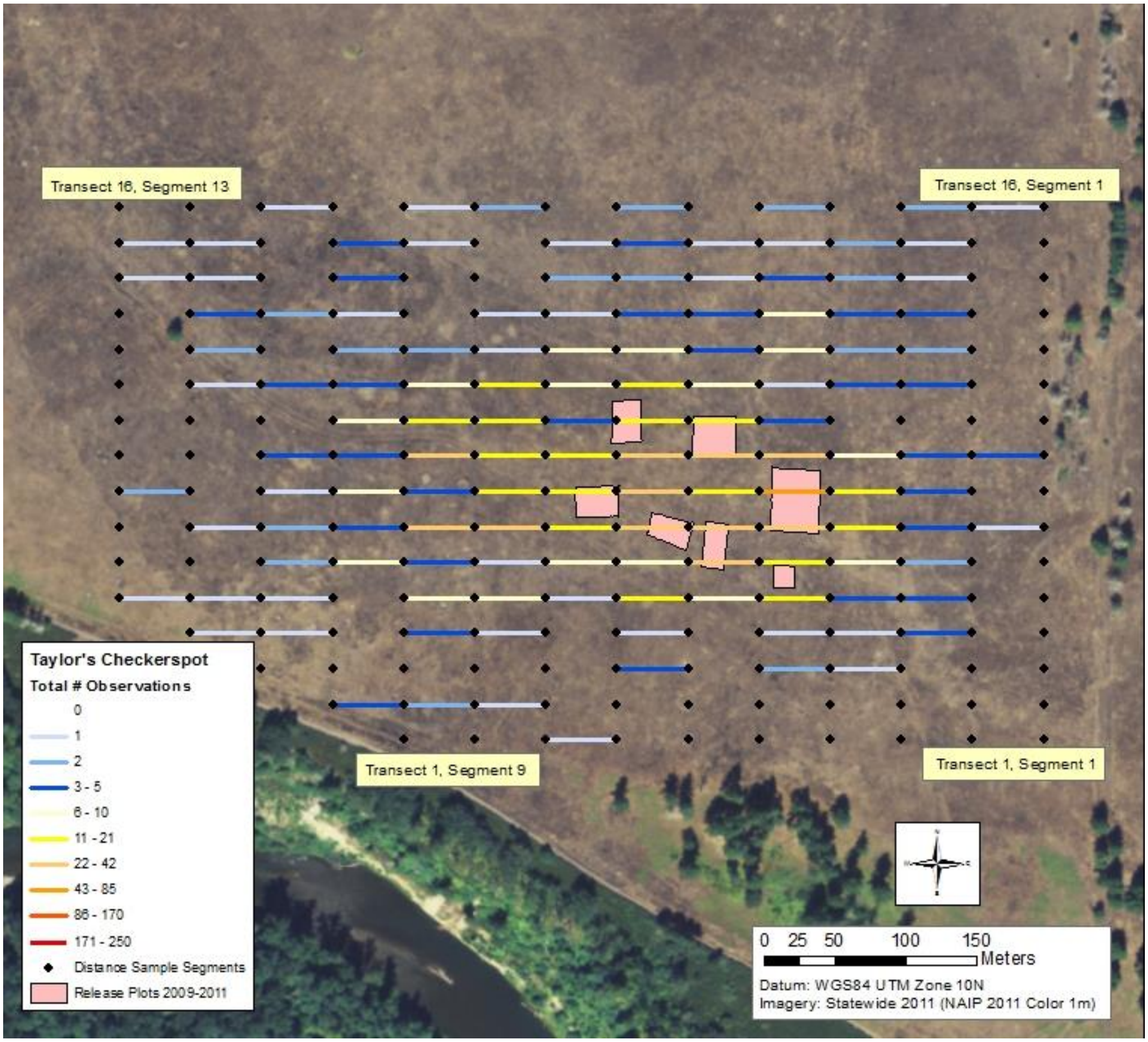


Fig. 2. Distribution of all adult Taylor's checkerspots observed during distance sampling surveys at Range 50, combined across all survey dates in Spring 2013, South Puget Sound, Washington.

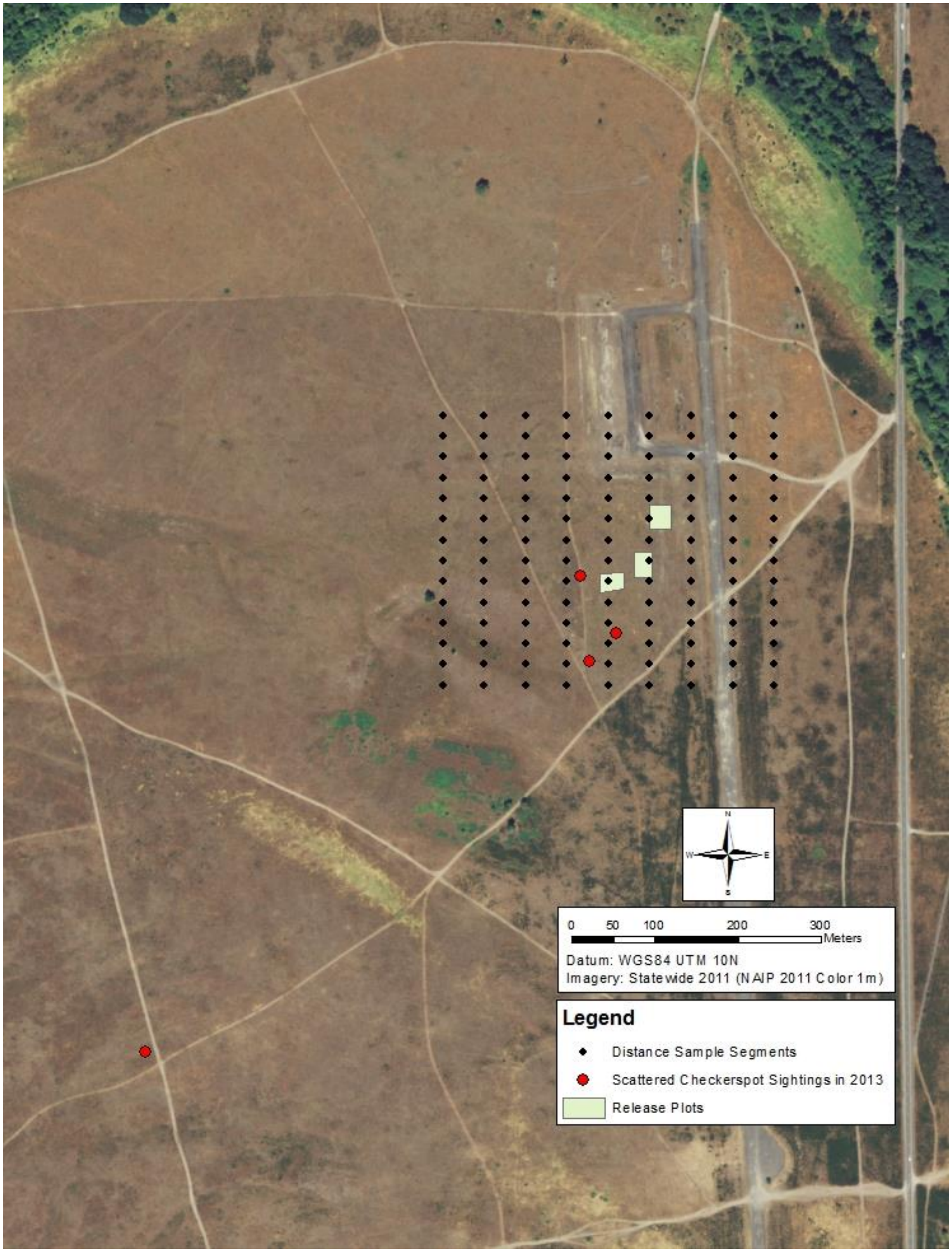


Fig. 3. Distribution of all adult Taylor's checkerspots observed at Pacemaker, combined across all dates in Spring 2013, South Puget Sound, Washington. No checkerspots were observed during distance sampling surveys, but a few were sighted on other occasions during the adult flight period.

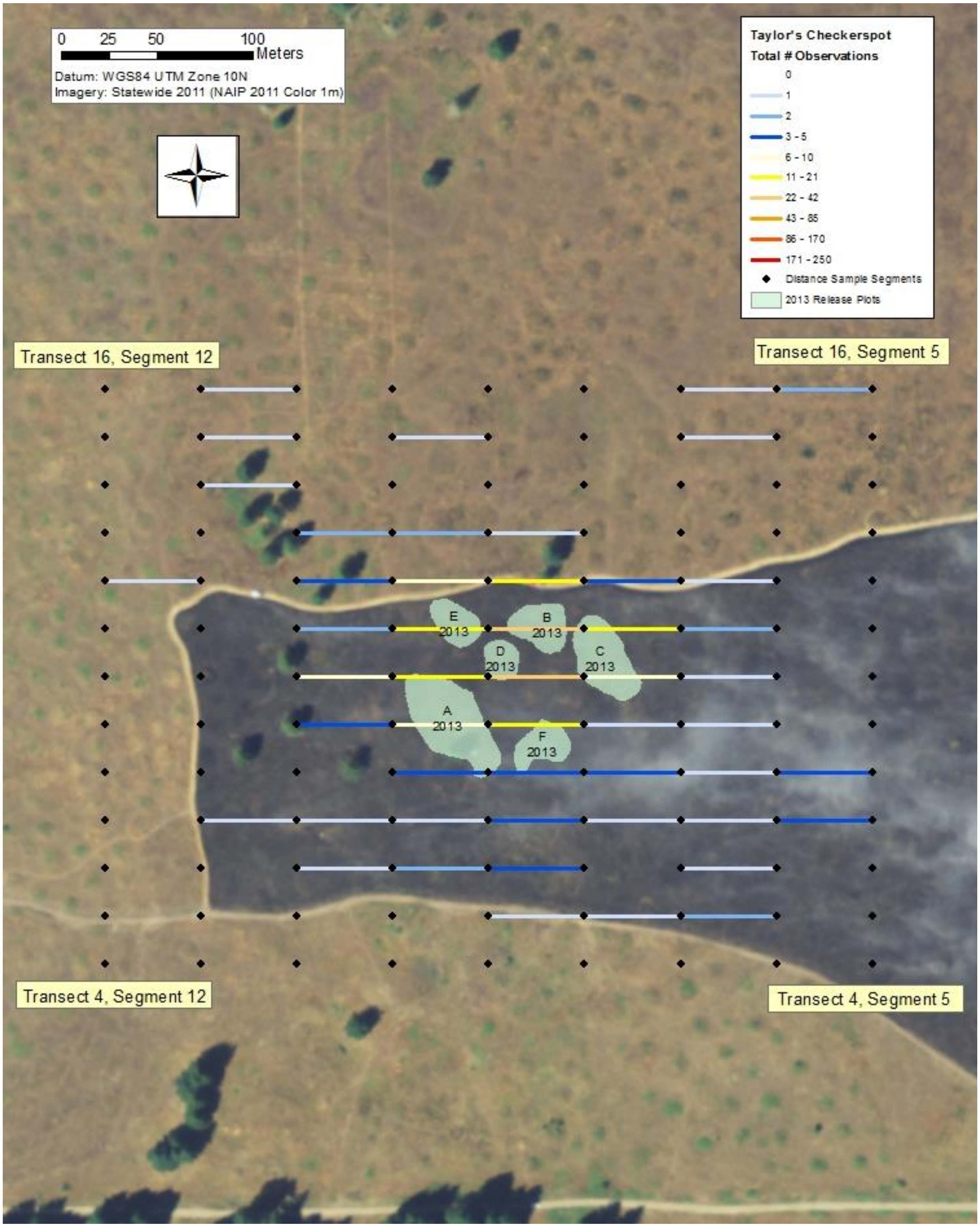


Fig. 4. Distribution of all adult Taylor's checkerspots observed during distance sampling surveys at Glacial Heritage Preserve, combined across all survey dates in Spring 2013, South Puget Sound, Washington.

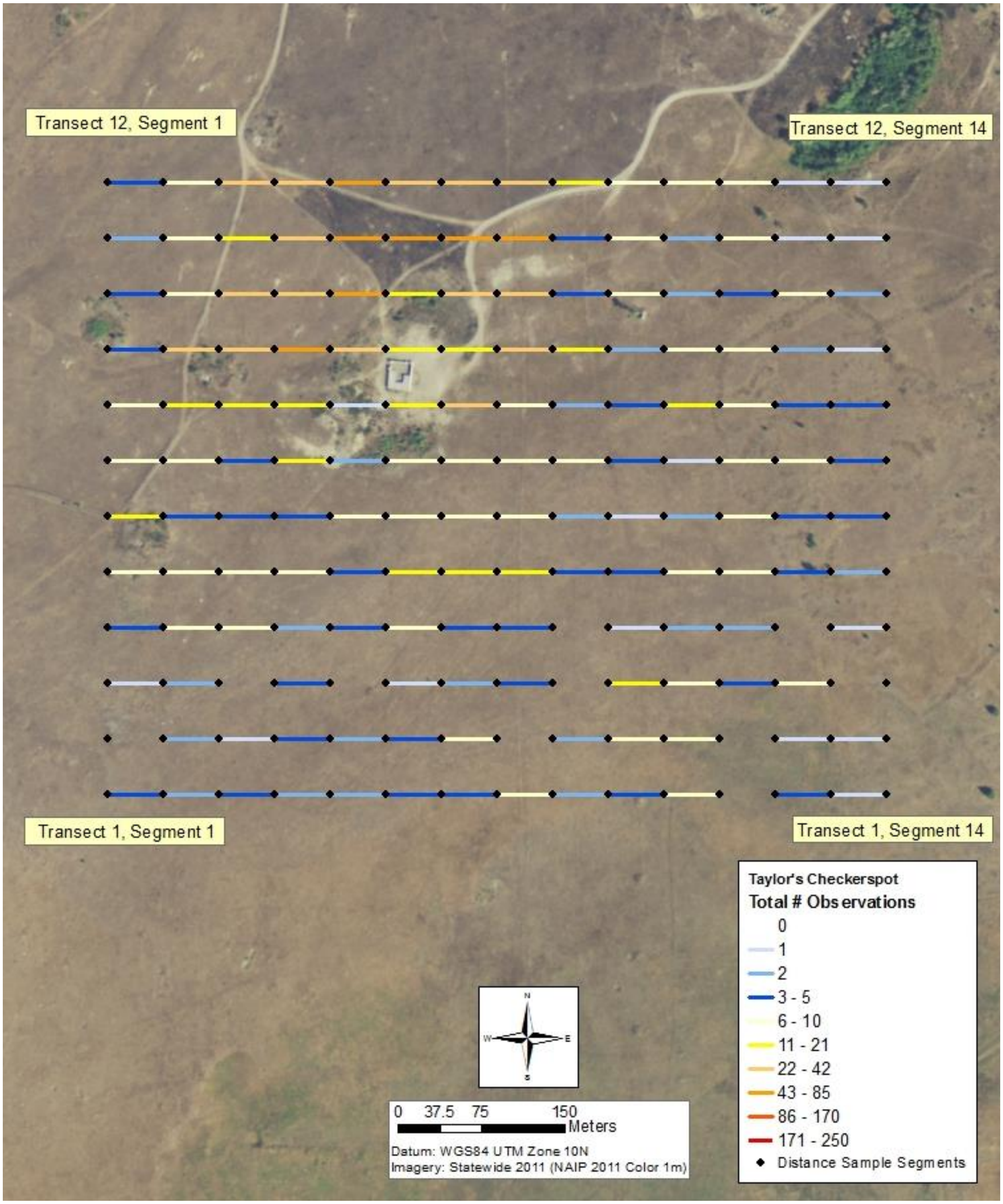


Fig. 5. Distribution of all adult Taylor's checkerspots observed during distance sampling surveys at Range 76, combined across all survey dates in Spring 2013, South Puget Sound, Washington.

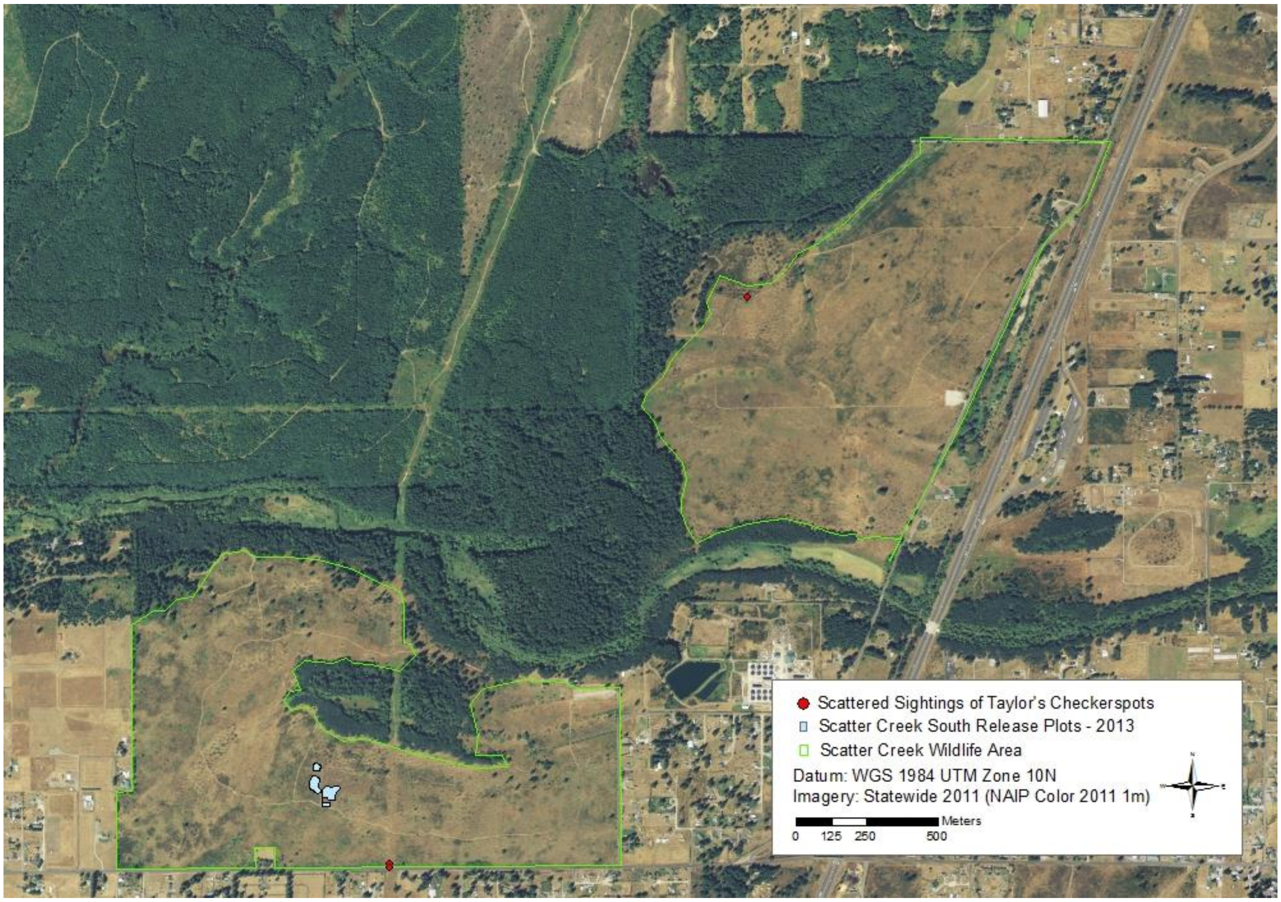


Fig. 6. Scattered sightings of adult Taylor's checkerspots observed at the Scatter Creek Wildlife Area, Spring 2013, South Puget Sound, Washington. Checkerspots have been observed flying across the road from the south edge of Scatter Creek's South Unit. The sighting north-east of the release plots in Scatter Creek's South Unit represents a new location since reintroductions began in 2007; checkerspots have not been observed at the North Unit since 1998 (Stinson 2005).

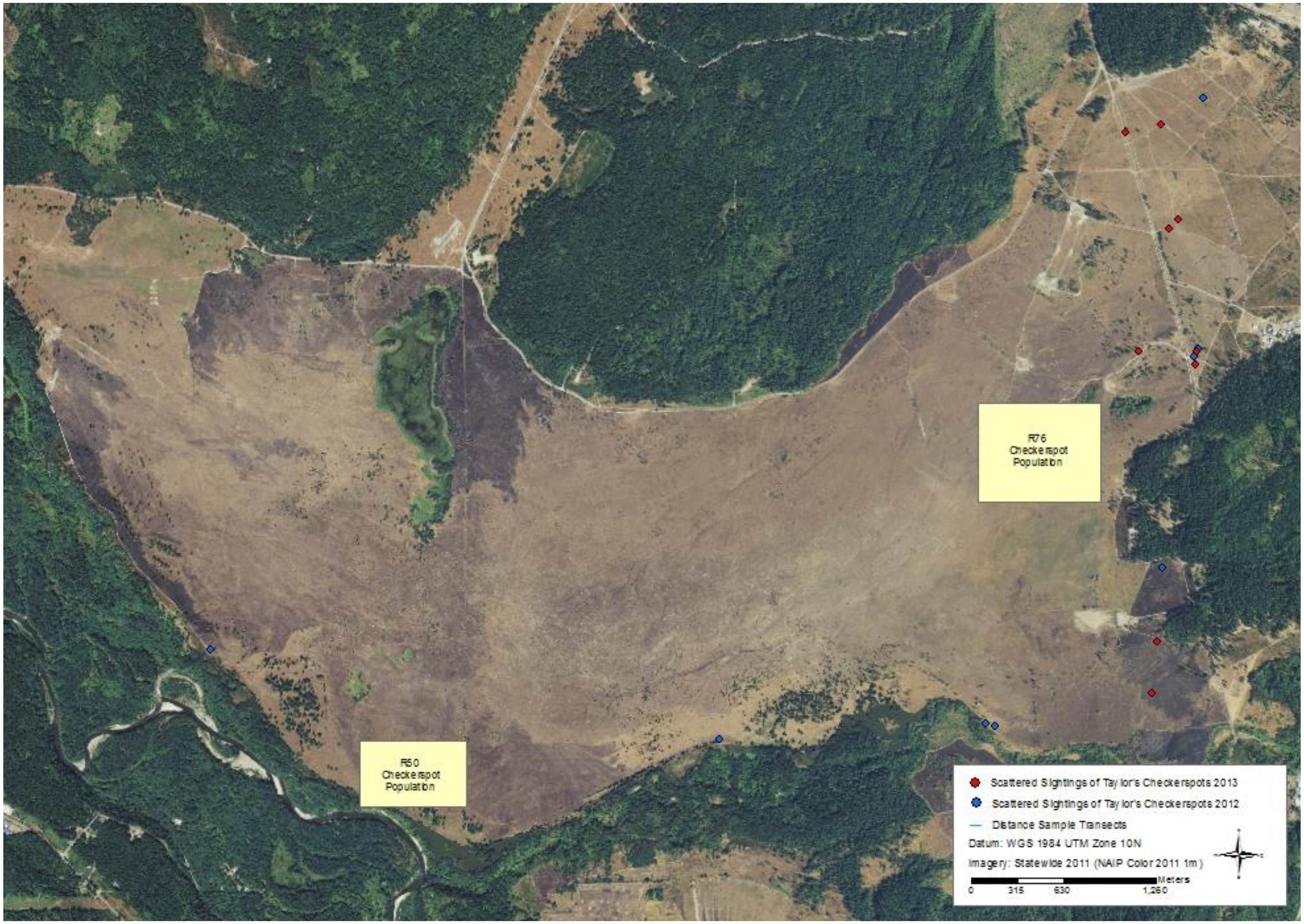


Fig. 7. Scattered sightings of adult Taylor's checkerspots observed around the perimeter of the Artillery Impact Area on Joint Base Lewis-McChord relative to survey areas at R76 (extant site) and the reintroduced population at R50, Spring 2012 and 2013, South Puget Sound, Washington. Sightings east and west of R50 represent new locations since monitoring began in 2003.