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ORIGINAL PAPER

Population size and conservation of the last eastern remnants of the regal fritillary, *Speyeria idalia* (Drury) [Lepidoptera, Nymphalidae]; implications for temperate grassland restoration

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Abstract We monitored the last remaining Pennsylvania population of the regal fritillary butterfly (*Speyeria idalia*) for 8 years (1997–2005) at Fort Indiantown Gap, a National Guard training facility located in south-central Pennsylvania, USA. We observed demes of this population in five grassland areas, four of which received limited protection from anthropogenic military activities (i.e., motorized vehicles were prohibited). The Pollard walk technique allowed us to make estimates of butterfly density over the consecutive 8 years period. Mark/Recapture estimates of population size were done in 2005. Of the three areas surveyed for relative abundance over the entire 8 years period, none showed significant changes in relative population density of adult butterflies, indicating that the population remained stable over the 8 years. Mark-recapture estimates of population size in 2005 indicate that this population reached a peak of 913 butterflies that year. The stability of population size occurred despite efforts to improve habitat. Dead caterpillars were found over 3 years in some fields indicating that caterpillar habitat includes both violets (caterpillar food plants) and mature warm season grasses approximately 5 years post-disturbance. These disturbances are caused by military activity such as motorized vehicle training that reduces areas to bare soil. These activities are now banned in protected butterfly areas.

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Introduction

The regal fritillary butterfly (*Speyeria idalia*) historically ranged from Nova Scotia south to northern Georgia and west to the Dakotas (Swengel 1993). Currently, only two extant populations have been documented east of Indiana (Derge and Chazal 2000; Williams 2001a). One of these populations, in south-central Pennsylvania, occurs within the boundaries of the National Guard Training Center at Fort Indiantown Gap (FTIG), Annville, Pennsylvania (Fig. 1). A recent genetic analyses suggested that this remnant population represents an eastern subspecies, *S. idalia idalia*, taxonomically distinct from the other remaining eastern population in Virginia as well as from remaining western populations (Derge and Chazal 2000; Williams 1999, 2001b; Williams et al. 2003); however, a more recent genetic study of the species determined that although the FTIG population is distinct from the population in Virginia, the evolutionary history of the species is not as straightforward as previously described and the designation of eastern and western subspecies may not be appropriate (Fonseca et al. 2008). Between 1998 and 2005, we monitored the one remaining Pennsylvania *S. idalia* population and examined its life history strategies. Over the 8-year period, two types of monitoring were used to gauge the size and stability of this population.

Habitat fragmentation and loss due to development, fire suppression, and inappropriate land stewardship, as well as pesticide misuse, have likely contributed to the decline or extirpation of butterfly populations of several species

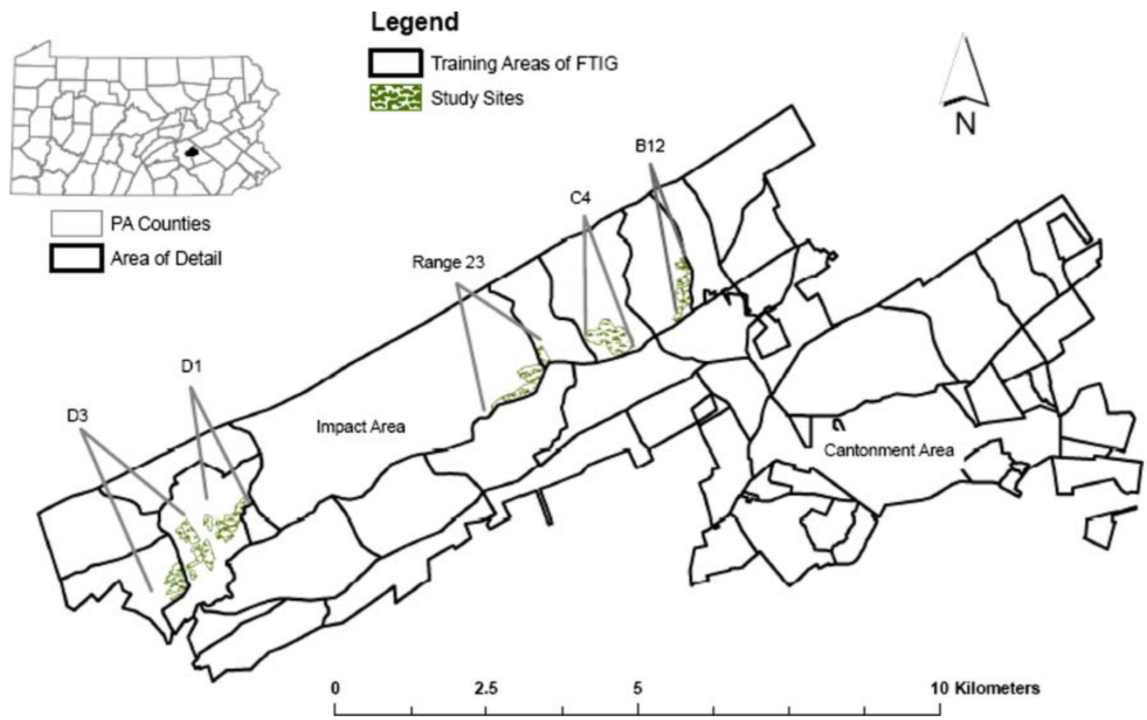

Fig. 1 Map of Fort Indiantown Gap training area. Monitored grassland butterfly sites D1, D3, Range 23, C4, and B12 are labeled within FTIG training areas

(Cushman and Murphy 1993; Longcore et al. 2003; Schultz and Crone 1998; Davies et al. 2005). Three federally endangered subspecies of *Speyeria zerene*, Myrtle's silverspot (*S. z. myrtleae*), the Oregon silverspot (*S. z. hippolyta*), and Behren's silverspot (*S. z. behrensii*), depend on violets as larval food (Opler and Warren 2002), as does the congeneric, yet currently unlisted, *S. idalia*. The eastern populations of the regal fritillary, a grassland specialist, are dependent on early successional, or disturbance maintained, plant communities. Host plants that *S. idalia* and other rare butterflies require as food sources often compete poorly with other plants (especially woody species). Without natural disturbance regimes such as physical soil disturbances (as occurred with large mammals now extinct in the NE United States [Bjorn and Anderson 1980; Trager et al. 2004]) or intermittent fire, these habitats and their butterflies disappear (Cushman and Murphy 1993; Schultz and Crone 1998; Pollick 2004). Higher rates of disturbance, such as yearly mowing, tilling, or the frequent use of herbicides may act to reduce plant diversity, discouraging perennial plants (like many nectar plant species) that take longer to establish. Where disturbance is lacking, woody plants eventually dominate and replace or suppress growth of herbaceous species that require full sun. Grassland conservation efforts in the northeastern US must include some form of disturbance, but the mechanisms of disturbance (mowing, burning, plowing) used may have dramatically different effects on biodiversity.

Some of the grassland areas inhabited by *S. idalia* at FTIG were protected from most military activities by a 2-year Memorandum Of Understanding (MOU) signed in August 1999 between the Pennsylvania Army National Guard (PAARNG) and The Nature Conservancy (TNC). This MOU provided TNC the opportunity to manage 158 acres (63.9 ha) of regal habitat and to monitor the butterfly population. The MOU expired in 2000, and a habitat management plan, outlined in a 2001 Environmental Impact Statement (EIS), continued to dictate the procedure for protecting these areas for butterfly conservation and monitoring. As a result, most of these areas are restricted to motorized vehicle use, and military training within these areas was largely absent over the 8-year monitoring period. Over time, in most areas with sufficient rainfall and soil, succession replaces grassland with small trees and shrubs which out-compete grassland plants (at FTIG *Schizachyrium scoparium* and *Solidago juncea* dominate much of the grassland habitat used by butterflies), including the nectar plants important to these butterflies (e. g., *Asclepias tuberosa*, *A. syriaca*, *Cirsium* spp., B. Ferster, unpublished data). All species of *Speyeria* feed on plants of *Viola* as caterpillars (Opler 1992). At FTIG, *Viola sagittata* (arrow leaved violet) is most abundant in grassland areas, and evidence of herbivory is common (B. Ferster, personnel observation). *Speyeria idalia* caterpillars hatch in late fall and overwinter as first instar caterpillars (Edwards 1879; Scudder 1889; Mattoon et al. 1971). In early spring the caterpillars become active, feed, and then pupate for approximately 2.5–4 weeks (Maynard 1886; Edwards 1879; Wagner et al. 1997). Typically, adult males emerge in mid-June (14–28 June at FTIG from 1998 to 2005, Fig. 2). Females emerge about 2 weeks later (29 June to 11 July at FTIG from 1998 to 2005, Fig. 2) each year (Scudder 1889; Denton 1900; Klots 1951; Opler 1983, 1992). Mating begins as soon as females begin to emerge and can be seen throughout the season. Males die not long after females emerge. Females spend much of the summer months in a reproductive diapause (Kopper et al. 2001), resting in shrubby vegetation or in tall grass. Although they can be seen nectaring on plants, females are more often counted along survey routes when flushed up from hiding places. Oviposition at FTIG begins in late summer after violets have senesced. Females deposit eggs in a seemingly haphazard fashion on the ground near areas that contain violets.

Here, we report on butterfly population density among years and among areas, and morphometric comparison of sexes and demes. We ask: (1) Is the overall butterfly population stable or changing? (2) Are there differences in population dynamics among the subpopulations? (3) Are there morphometric differences among the subpopulations? And; (4) What are the habitat preferences of the caterpillar population?

The intent of the population monitoring of this species was to determine whether the population was increasing, declining, or stable. The subpopulations may exhibit different dynamics to the population as a whole and each could influence the overall trend. We were interested to determine if there was an overall population effect and if any of the subpopulations were contributing disproportionately to that effect. The results would then be valuable for future management efforts through more precisely targeted efforts. Morphometric analysis allowed us to investigate the possible isolation of subpopulations from each other, as suggested by recent genetic studies (Keyghobadi et al. 2006). If subpopulations were found to be significantly different in body-size measurements we might suspect that subpopulations have not interbred for some time. We discuss the interpretation of these data, compare the results with recent genetic studies, and examine the implications for conservation practices of this rare and declining species.

Methods

Transect survey routes

We established survey routes using the Pollard Walk Technique (Pollard and Yates 1993) in 1998, 1999, and 2002 (Table 1) to estimate relative abundance through five grassland areas within FTIG where *S. idalia* adults were seen regularly over the season of activity. Four of the five areas were part of the original MOU, and these areas continued to be protected from most military activities that might disturb habitat after the MOU expired. We established routes through areas that ranged from 14 to 33 ha. Transect lines ranged from 1.9 to 3.4 km in length (Table 1). Transects varied in length, orientation, and habitat type traversed (e.g., forest, grassland, riparian). Transect designers attempted to maximize sampling within

Fig. 2 Phenology of Regal Fritillary at FITG from 1998 to 2005. *Y*-axis units are days on the Ordinal calendar. *Light gray bars* denote the time from emergence of first male to the emergence of the first female, *dark gray bars* are the flight periods of the females, and *black diamond* is the estimated day of peak emergence for the population from the program INCA

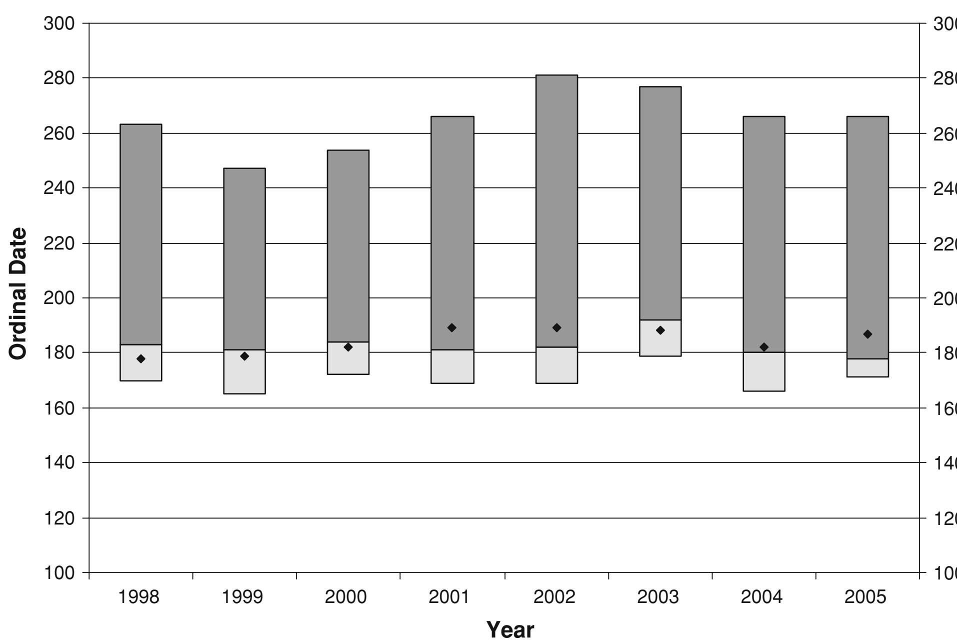

Table 1 Length and size of permanent transect routes established at Fort Indiantown Gap, Annville, PA for monitoring of Regal fritillary butterflies over 8 years

Route location	Length of transect (km)	Year established	Area of MOU		Area surveyed (km^2) = length × 40 m
			Acreage	Hectares	
B12	1.9	1998	33.7	13.64	0.069
D3	3.4	1998	82.4	33.35	0.124
Range 23	2.1	1998	54.7	22.14	0.076
C4	1.6	1999	N/A	N/A	0.058
D1	1.9	2002	49	19.83	0.069

N/A Not available

the habitat mosaic of each area (TNC 2001). Transects therefore do not sample the same size areas of any given habitat type (e.g., forest, grassland, riparian etc.) within areas. We compensated for this bias by estimating population density using an index of population size (the Insect Count Analyzer, INCA [INCA 2002; Longcore et al. 2003; Haddad et al. 2008]) divided by the area surveyed (Rabasa et al. 2007). We began walking transects the first week congeneric great spangled fritillaries (*Speyeria cybele*) were seen, and continued walking weekly transects at each site until a week after the last regal adult was seen, 13 to 16 weeks total for each year. A single observer walked transects at a steady pace (mean = 3.02 ± 0.29 km/hr) between 10:00 a.m. and 3:00 p.m., when wind speed was low (<10mph) and temperatures were between 23 and 37°C. Before walking each transect, we recorded time, temperature, percent cloud cover, and wind speed (using Beaufort scale). Observers then moved between 1.8 m tall wooden-stake markers along the route using compass bearings marked on the white-painted tops. As we walked, observers scanned an area of approximately 18.3 m on either side of the transect line, so that a 35 m wide band was observed along the route. We recorded the following data for each butterfly sighting: the section of transect, behavior (flying, nectaring, flushed, resting, etc.), gender, and the nectar-plant species on which nectaring butterflies were seen feeding. Because of the clear visibility of this large butterfly species in grassland habitat, we assumed that all butterflies within the 35 m width of the transect were counted (Harker and Shreeve 2008). Time and temperature at the end of a session were also recorded. Weekly surveys began when the first males were observed in the field, and ended 1 week after the last female butterflies were seen flying. Occasionally, in some years, researchers were not allowed access to sites during weeks when scheduled military training was heavy. On a few occasions, particular routes were not walked during a week when weather conditions did not meet the minimum requirements above. In these instances, means of the weeks previous and following were substituted for missing data. The study sites include five open, grassland habitat sites (B12, Range 23, C4, D1, and D3) (Fig. 1) within a forested landscape dominated by oak and maple (Latham et al. 2007; TNC 2000). These areas are designated as training areas of the Fort Indiantown Gap facility, and may be connected by the "impact zone" between them. However, this area is was off-limits to research due to the possibility of unexploded objects in the area. Mark-recapture allowed us to evaluate the connectivity of sites.

Mark recapture

We captured butterflies by long-handled aerial net (15" diameter, Bioquip products, Dominquez, CA) when encountered, July 11–15 and 25–29, 2005. We marked individuals with unique numbers on the front and back of the right forewing, using a black, fine-point Sharpie® permanent marker pen. Individual information was recorded along with area, field, and time of capture. Body length (including head to posterior abdominal segment), wing length (from R_1 at outer wing margin to C at inner wing margin articulation with mesothorax), and head width including eye, were measured for each newly captured individual to the nearest .01 mm with an electronic digital calipers (0–150 mm Whitworth), or a Swiss Precision dial caliper (Spi 2000 6"/0.001"). Butterflies were captured, marked and measured, and then released. Fields were revisited 2 or 3 days later for recapture. Approximately equal numbers of person-hours were spent in each field in order to standardize effort. The mark/recapture was done in 11 days on two nonconsecutive weeks because military activities prevented access to sites during the intervening week.

Caterpillar surveys

In 2001–2005 we searched for caterpillars by sitting on the ground and looking, first for evidence of herbivory on violet leaves and then carefully searching the ground around the partially eaten violets and in the grass surrounding them for caterpillars.

In 2002 monitors began to find dead *Speyeria* sp. caterpillars attached to the tops of dead grass stalks and limbs of small trees in one particular field (M. T. Swartz, personnel communication). In 2003 we searched for dead caterpillars daily as soon as the first one of the season was discovered. Dead caterpillar searches ended only when no dead caterpillars had been found for one full week. Caterpillar location was marked using Trimble Total Station 5700 GPS unit (Trimble Navigation Ltd., Sunnyvale CA) and Arc GIS 9.0 (ArcGIS 9.0 ESRI GIS and Mapping Software, Redlands CA). Each caterpillar was collected in a clean, glass vial, head width and body length were measured, and caterpillars were frozen for future analysis for pathogens.

Statistical tests

Pollard walk

We assumed that each area surveyed was independent of the others. This assumption is justified by genetic analysis demonstrating that gene flow is restricted to small local areas in this species (Keyghobadi et al. 2006). That study also determined that two areas were genetically similar (D1 and D3), but because D1 was not surveyed through all 8 years (Table 1), we used data from D3 for these analyses. We used an index of abundance (INCA) converted to density to compare trends over time at each area. We did not include Area C4 in the comparison because of the small population size. We took the log transformed density index and calculated linear regressions of B12, D3, R23 and the total of all sites to determine if populations were increasing, decreasing or staying stable (Roy and Sparks 2000). We used One-way ANOVA with Tukey post hoc comparisons to determine which areas differed from the others in average density (Zar 1999). INCA population estimates also yield an estimate of peak emergence time. We examined this estimate along with the time of first male emergence, first female emergence, and last female sighted. Statistical analyses were performed with SPSS version 12.01 or Minitab version 15.1.1.

Mark-recapture

Jolly-Seber model A for open populations (Jolly 1965) was used to estimate population sizes over four time intervals for males and females separately. Jolly-Seber is designed for open populations, allowing births, deaths and immigration (Krebs 1999). Males and females were pooled for total population sizes at the three sites as Chapman's Test (Chapman 1951) analysis indicated no difference in catchability between males and females. In this analysis, we included B12, D1 and D3 together as D, and Range 23; Fig. 1. These three subpopulations are connected only by rare instances of dispersal and gene flow (Keyghobadi et al. 2006). Jolly-Seber estimates of probability of survival and probability of capture were calculated using Program Mark (Cooch and White 2006), and Maximum Likelihood Estimators were used to find the best fit model (Table 2).

Morphometrics

We measured wing length, total body length, and head length to the nearest 0.01 mm with an electronic digital calipers (0–150 mm Whitworth), or a Swiss Precision dial caliper (Spi 2000 6"/0.001"). Because of the conservation status of this species and the risk of injury, we did not weigh the individuals. As a rough measure of wing loading, we calculated the ratio of wing length to body length. Heavier individuals would be expected to have a longer body length. This index would be proportional to a measure of wing loading (weight/length across both wings and thorax).

We used General Linear Model (GLM) to examine the differences among sites and sex as fixed factors and the interaction of these factors for all four variables. We used Holm–Sidak post hoc tests to determine the groups that differed significantly in pairwise multiple comparisons.

Results

Population survey

Mean density of butterflies in the three areas (B12, D3, and R23) was 502.51/km^2 (SD = 73.51). Average density (SD in parentheses) in separate areas were: B12 = 729.35 (338.93), D3 = 420.77 (171.80), R23 = 429.93 (166.24). The density of butterflies for the entire survey site did not differ significantly over 8 years ($F = 0.55$; df = 6;

Table 2 Jolly-Seber estimates of population sizes for three areas

Jolly-Seber estimates Size of population (N)						
Collection #	B12-males	B12-females	D-males	D-females	R23-males	R23-females
1	74.67	41.50	209.33	84.79	189.89	312.86
2	61.71	48.23	86.92	257.87	94.84	52.20
3	30.48	42.42	33.06	159.31	15.30	40.32
4	21.65	15.30	109.77	167.32	100.01	135.13

Population estimates were calculated for four periods during the flight season

$P = 0.49$; Fig. 3), however, density in B12 was significantly greater than D3 and R23 ($F = 4.84$, df = 2, $P = 0.02$; Fig. 4). One subpopulation or deme (C4) remained significantly smaller than the other demes throughout the monitoring period and we did not include it in the analysis. Regressions of the trends of log index of the population density over the 8 years were not significant (B12: $F = 3.32$, df = 1, $P = 0.12$; D3: $F = 2.89$, df = 1, $P = 0.14$; R23: $F = 0.66$, df = 1, $P = 0.45$; Total: $F = 0.55$, df = 1, $P = 0.49$).

We examined the phenology of *S. idalia* (Fig. 2). The INCA estimate of the time of peak emergence varied across the years, but not significantly (Peak emergence over years: $F = 1.10$, df = 2, $P = 0.35$).

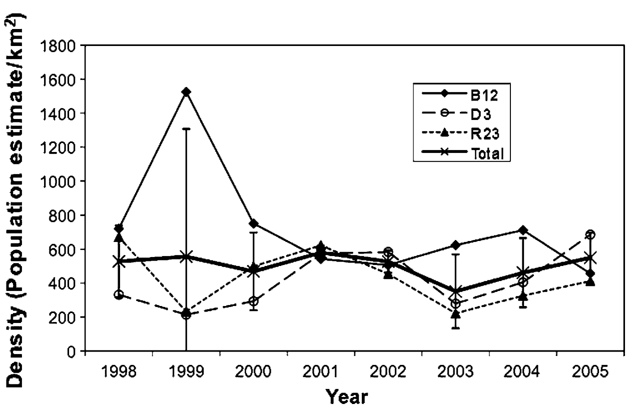

Fig. 3 Population estimates for each year from 1998 to 2005 generated by the program INCA (INCA 2002; Zonneveld 1991). Three subpopulations are displayed: B12, D3, and R23, plus the total for the metapopulation. C4 did not have sufficient samples from each year to generate estimates. The line for total population for the site includes *error bars* (±1 SD)

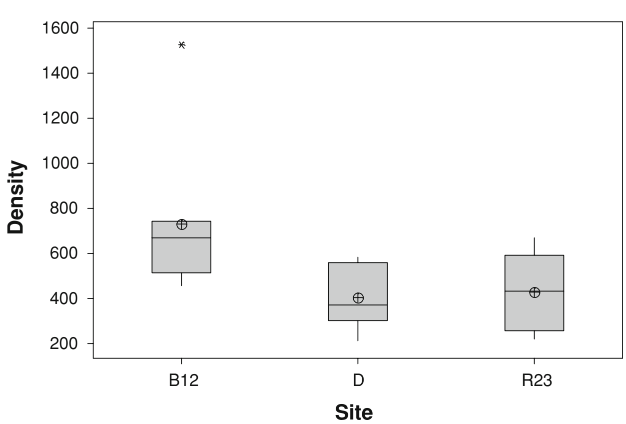

Fig. 4 Density of butterflies at each site over 8 years. Boxes show 25th, 50th (central horizontal line), and 75th quartiles. *Whiskers* delineate the range, *asterisk* the outlier, and *circle* with *plus symbol* the mean

Mark recapture

Six hundred and thirty-three individual butterflies were captured and marked over the 2-week capture period in 2005. Of those captured, 273 were female, and 360 were male. Forty-three percent of these individuals were recaptured at least once. A total of 11 butterflies were documented to disperse between different areas. Two marked females and three marked males moved between B12 and D. Four marked males and a single marked female moved between B12 and Range 23. One marked female but no marked males moved between D and Range 23. Five butterflies were seen dispersing between B12 and C4, but these movements are not counted as dispersal due to the proximity of the two areas.

Males and females were pooled for total population sizes at the three sites as Chapman's Test (Chapman 1951) analysis indicated no difference between males and females in catchability ($X^2 = 6.00$, df = 4, $P = 0.199$). Jolly-Seber yielded four estimates of population size over the season (Amstrup et al. 2005). For insect populations that change considerably from week to week, as butterflies do, Jolly-Seber estimates tend to be more precise than Peterson's, which only estimates populations between two time periods and not continuously (Krebs 1999).

Jolly-Seber estimates of probability of survival and probability of capture were calculated using Program Mark (Cooch and White 2006), and Maximum Likelihood Estimators indicate the best fit model (AICc weight = 0.875) for probability of survivorship and probability of capture includes time dependency for probability for survival and group (sex and area) dependency for probability of capture. The models $\Phi(t)$ $p(g \times t)$ and $\Phi(g \times t)$ $p(g)$ (where Φ = probability of survival, p = probability of capture, t = time, g = area and sex) both fit the data reasonably well ($X^2 = 3.350$, df = 4, $P = 0.5011$, effective sample size = 640). Jolly-Seber estimates of population size at each time interval were different among sites and between sexes. Thus, subpopulations of adult butterflies at FTIG differ in size over the season and male and female populations fluctuate, asynchronously.

Morphometrics

Females tend to be larger than males (Howe 1975). Females had longer wings than males at sites D and Range 23, but at site B12 females were smaller than males. Females at B12 had the smallest wings and the smallest wing-to-body ratio (Table 3) of any site, and males at B12 were significantly larger than males at D or Range 23 in wing length. Head width and body size did not differ significantly among the sites within each sex. There were only 13 individuals of each sex for which morphometric data

Table 3 Mean (SD) of four body measurements for males and females at the three sites

Sex	Site	N	Wing	Body	Head	Wing-body ratio
F	B12	13	40.62 (3.68)	30.63 (2.00)	5.26 (0.46)	1.33 (0.14)
F	D	75	45.19 (2.39)	30.70 (1.45)	5.14 (0.28)	1.47 (0.07)
F	Range 23	55	45.60 (2.32)	30.53 (1.56)	5.16 (0.31)	1.50 (0.07)
M	B12	13	41.92 (3.09)	29.28 (1.49)	5.31 (0.24)	1.43 (0.12)
M	D	94	39.61 (1.78)	29.43 (1.78)	5.27 (0.28)	1.35 (0.07)
M	Range 23	131	40.11 (2.17)	29.68 (1.89)	5.17 (0.50)	1.35 (0.08)

could be analyzed in B12; perhaps the differences we observed are a result of small sample size. However, because head width and body size were not significantly different between B12 and either Range 23 or D, these wing length differences may not be a result of sampling error. We found wing size/body size ratio to be significantly lower in B12, than for other sites which indicates that it is only wing size that has changed in the B12 deme, not body size (Table 4).

Table 4 ANOVA table of morphometric measurements of males and females at three areas (B12, D, R23). (a) ANOVA table; (b) Multiple comparisons from Holm-Sidak method (B = B12, R = R23)

Variable	Source	*df*	*F*	*P*
ANOVA table				
Wing	Sex	1	92.51	<0.001
	Site	2	6.02	0.003
	Sex × site	2	27.83	<0.001
	Residual	375		
Body	Sex	1	20.85	<0.001
	Site	2	0.10	0.907
	Sex × site	2	0.72	0.486
	Residual	375		
Head	Sex	1	1.29	0.257
	Site	2	1.38	0.252
	Sex × site	2	0.99	0.373
	Residual	375		
Wing/body ratio	Sex	1	19.16	<0.001
	Site	2	3.43	0.033
	Sex × site	2	25.43	<0.001
	Residual	375		

Variable	Sex	Site
Pairwise multiple comparisons		
Wing overall	<0.001	<0.001
R vs. B	<0.001	<0.001
D vs. B	<0.001	0.006
R vs. D	0.296	0.097
Body overall	<0.001	NS
R vs. B	NS	NS
D vs. B	NS	NS
R vs. D	NS	NS
Head overall	NS	NS
R vs. B	NS	NS
D vs. B	NS	NS
R vs. D	NS	NS
Wing/body overall	<0.001	0.033
R vs. B	<0.001	0.015
D vs. B	<0.001	0.101
R vs. D	0.125	0.121

Caterpillar surveys

In 1998, nine *Speyeria* caterpillars were found by placing 2.5 cm^2 squares of white sheet on the ground overnight. However, in 1999 and 2000 no caterpillars were found using this method. In 2001 we searched for caterpillars by sitting on the ground and looking for evidence of herbivory on violets before more thoroughly searching the ground nearby. This method also did not result in many found caterpillars. In 5 years we found only 13 *S. idalia* after approximately 300 observer-hours of searching in all five sites.

We first found dead *Speyeria* sp. caterpillars stuck to the tops of dead grass stalks and limbs of small trees in 2002. They were neither collected nor counted. In 2003, we found 116 dead *Speyeria* sp. caterpillars. In 2004 we found a total of 287 dead *Speyeria* sp. caterpillars in four different areas. In 2005 we found 51 dead *Speyeria* sp. caterpillars. Too few of each cohort was positively identified as *S. idalia* to analyze differences among areas statistically. We overlaid dead caterpillar locations onto vegetation maps, and examined records of disturbance in these areas for possible correlations (Fig. 5).

During the 2005 field season a pupa was also discovered. To our knowledge it is the first time this life stage has ever been found in the wild (Fig. 6). This pupa was attached by its cremaster hook to the underside of a warm season grass tussock, approximately 1 cm above the ground. We placed a cage over the tussock in the hopes of capturing and marking the newly eclosed adult, but were unsuccessful. At the end of the season, when we removed the cage, we found the pupal skin, open along the sutures.

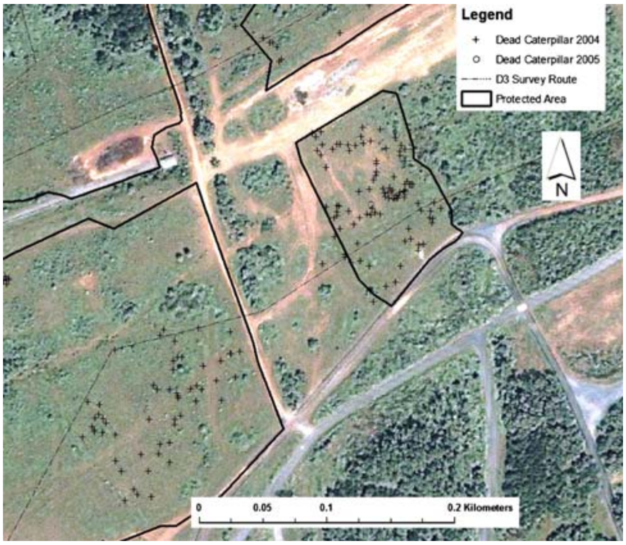

Fig. 5 Map of dead caterpillars found in 2005 in a field of area D where motorized vehicle maneuvering practice by military personnel had caused disturbance 5 years earlier. *Dots* are individuals dead caterpillars. *Blue line* is the survey route for regal fritillary adults. *Black lines* show boundary of areas protected for butterflies

Fig. 6 Photograph of regal fritillary pupa attached to the underside of grass tussock. Photo Dave Zapotok

We assume the adult eclosed, crawled out of the tussock, and out of the cage before spreading its wings for drying and then flying away, unmarked.

Discussion

Butterfly populations naturally fluctuate over years, and this is also true of the FTIG Eastern regal fritillary population. However, in only one case are these differences significant. In one area (B12) the number of butterflies counted along a permanent transect was significantly higher in 1999 than it was along that same route in other years. In no other site were significant differences found among years. Over the 8-year period the standard deviation from the mean had not increased (Fig. 3). The variation in population density over time (Fig. 3) is likely to have a greater impact on the persistence of the population than size (Kuefler et al. 2008). The probability of metapopulation extinction is the product of the probability of extinction combined over the subpopulations (Morris and Doak 2002). The continued existence of the subpopulations becomes important for metapopulation sustainability. Large fluctuations in subpopulation sizes put the larger metapopulation at risk (Matern et al. 2007).

Mark-recapture population size estimates indicate that the population was approximately 913 individuals at its peak in 2005, in three demes with little migration between them. Subpopulation sizes of adult butterflies at the FTIG site differ over the season and male and female populations fluctuate, but not synchronously. Although the population appeared to be stable up through 2005 (Fig. 3), reliance on one metapopulation of relatively small effective size to insure population persistence is risky.

Morphometric analysis of wing length, body length and head width found B12 to be unusual in having smaller winged females and larger winged males, while the other populations have smaller males. Keyghobadi et al. (2006) found no genetic differences between B12 and Range 23, but found that these two demes differed from those at sites D1 and D3 (which did not differ from each other) in allele frequency. B12 is geographically closer to Range 23 than either is to D, although Range 23 and D are connected to each other by the open and frequently burned impact zone (Fig. 1). The mark/recapture data indicate that migration among sites is low. Genetic work supports these findings (Keyghobadi et al. 2006). Low migration among demes might protect the population from spread of disease, but also means that these small demes area at a greater risk of extinction, due to inbreeding and low heterogeneity (Frankham and Ralls 1998; Saccheri et al. 1998).

Butterflies in area C4 may represent immigrants from a nearby, stable population (B12), rather than resident butterflies. These two sites are separated by an artificial pond, a road, and a narrow band of red maple forest. The C4 grassland site likely represents marginal habitat unsuitable for supporting a viable regal fritillary population.

The large, conspicuous adults are difficult to miss as they flutter about nectar plants or search the vegetation for mates or egg-laying sites. However, caterpillars have much more cryptic behavior and are difficult to find. Conservation efforts for only one life stage may be futile if another life stage is ignored. The dead and moribund caterpillars we found provide insight into the habitat requirements of this life history stage. When we examine maps where the location of dead caterpillars is overlaid onto aerial photographs we can see a clumped distribution of caterpillars in areas we can then examine for vegetation characteristics that may indicate habitat requirements for this cryptic life stage (Fig. 5). The area in D3 where most of the dead caterpillars were found in 2005 and the area in C4 where most of the dead caterpillars were found in 2004 are both areas that had been physically disturbed approximately 5 years earlier by mechanized vehicles. The C4 site is one that is not protected under the MOU, and one that consistently has a very small population of adult butterflies, possibly immigrants from nearby, protected B12 area. In both areas the mechanical disturbance resulted in bare, churned up soils that were then left largely alone to recover. Interestingly, such disturbance can increase violet density significantly for approximately a year before density is reduced to those found in undisturbed grassland (Latham et al. 2007). It is reasonable to assume that caterpillars are, therefore, not concentrated in areas of highest violet density, but are instead in areas where some factor other than the abundance of food plants is important. Warm-season grasses take longer to recover from disturbance (Latham et al. 2007) than violets do, and form tussocks of old leaves after a number of years. It was in one warm-season grass tussock that the hanging pupae was found (Fig. 6), and indeed caterpillars were sometimes found under these thick bunches of dead grass leaves. We suggest that the tussocks themselves provide a resource, perhaps protection from predators or provision of microenvironmental elements that caterpillars require. Conservation stewardship must therefore take into consideration the healthy growth of warm-season grasses as well as the density of the caterpillar food plants.

Butterflies faced with habitat of declining quality likely move on to other, more productive areas (Hanski 1999). Metapopulation theory predicts that if no suitable areas are available, populations will vanish (Hanski et al. 1994; Hanski 1999; Davies et al. 2005). At FTIG butterflies may be able to move to better habitat, but research efforts to understand habitat and butterfly activity, and stewardship activities designed to maintain habitat quality were restricted to the designated MOU areas. Stewardship activities in regal habitat at FTIG included prescribed burning and removal of woody vegetation by various mechanical methods (mowing, tree cutting). Statistical comparisons of areas where stewardship activities have been concentrated are problematic because no true controls or independent replicates exist. Recent literature has disputed the impact of fire on butterfly populations (Swengel 1996, 2001; Panzer 2002). Swengel (2001) reviewed the literature on the fire effects on insects populations and demonstrated that more research was needed. Before widespread prescribed burning becomes common practice for conservation of open areas, better understanding of the effects of fire on such communities is called for. Panzer (2002) found that only a small proportion of the remnant-dependant insect species he examined responded negatively to fire. Most species recovered within 1 year, and the remaining species recovered in 2 years or sooner.

The population of *S. idalia* at FTIG remained stable over the 8-years monitoring period despite these attempts at habitat improvement. It is possible that the carrying capacity has not been increased by stewardship in these habitats, or these stewardship activities may have kept populations from declining in monitored areas. At this point, we do not have enough data to determine if stewardship activities have kept the populations at FTIG from declining, or if the banning of activities that occasionally disturb areas within flight range of ovipositing females are maintaining suitable caterpillar habitat, and thus benefiting the population.

The FTIG population of *S. idalia* is one of two remaining eastern populations of a once common species. Populations at the northeastern edge of their range began disappearing by the mid 1950s, and extirpations progressed southward (Schweitzer 1984). To that end, Schweitzer (1993, 2000) noted a drastic decline in populations since 1980, where an increase in extirpations rates was seen after 1987. The species is considered extirpated or historical (SX/SH) from Maine, Massachusetts, New Hampshire, Vermont, New York, Connecticut, Rhode Island, New Jersey, Delaware, Maryland, West Virginia, Kentucky, Georgia, Ohio, and Indiana (PABS www.dickinson.edu/prorg/pabs/invertebrates.htm). Furthermore, *S. idalia* is now rapidly declining in the prairie states of Illinois, Iowa and Wisconsin (Debinski and Kelly 1998; Schlicht and Orwig 1998; West 1998; Williams 1999). Currently, this species has no federal protection status, although it is considered endangered by the Pennsylvania Biological Survey (PABS www.dickinson.edu/prorg/pabs/invertebrates.htm).

Speyeria idalia is a grassland specialist (Swengel 1998). Regal-occupied habitat at FTIG is old-field grassland dominated by native warm-season grasses and containing diverse forbs and some small, sparse, shrubby vegetation (Ferster et al. 2008). Fields inhabited by *S. idalia* at FTIG contain an average of 1.79 violets per m^2 (*Viola* spp., *V. sagittata* at FTIG) (B. Ferster, unpublished data), the caterpillar food plant, and a continual and reliable supply of nectar plants throughout the adult flight period. *Speyeria idalia* is a nectar generalist, but selective in its nectar feeding at FTIG. Nectar plants used at FTIG include common milkweed (*Asclepias syriaca*), butterfly milkweed (*A. tuberosa*), swamp milkweed (*A. incarnata*), pasture thistle (*Cirsium pumilum*), field thistle (*Cirsium discolor*), and wild bergamot (*Monarda fistulosa*). Mature warm season bunch grasses (e.g., little bluestem, *Schizachyrium scoparium*) provide protection for caterpillars and pupae (Ferster 2005). Small trees and shrubs provide resting sites for females during their long summer diapause. This habitat mosaic includes plant species that require periodic disturbance to reduce competition from woody vegetation (e.g., violets), as well as perennial species that do not always flower during their first year (e.g., milkweeds and thistles). Too frequent a disturbance would reduce the diversity of the plant community required by this charismatic butterfly species. Changes in land use practices across the historic range of this butterfly likely reduced plant diversity resulting in local butterfly extinctions. Fort Indiantown Gap's unusual land use practices have supported a population of *S. idalia* as well as a plant community that may also be rare. The frequency and intensity of disturbance at this military installation may mimic mechanisms of disturbance with which this grassland plant community and this butterfly evolved. Large track vehicle training practice may be a surrogate for the activities of megafauna that are no longer extant in the Northeastern United States (as "iron bison") (Bjorn and Anderson 1980; Trager et al. 2004). The mechanical soil disturbance these vehicles produce leads to a short-term increase in violet density (Latham et al. 2007). Nectar plants and tussock forming, warm-season grasses are slower to recover from such disturbances (Latham et al. 2007). As soon as butterfly areas were identified at FTIG, they were protected from motorized activity, and track vehicle training ended in those areas. The stewardship activities performed during the 8-years period of population monitoring likely do not mimic the iron or the real bison in disturbance intensity. The unchanging population at FTIG may indicate a slow deterioration of their grassland habitat, as a result of decreasing plant diversity, perhaps a result of inappropriate stewardship activities. Because this butterfly population represents unique biodiversity of the last vestiges of a species that survived in suitable patches throughout the eastern US until our land use practices were altered, it would be prudent to examine conservation and land management practices used to maintain open "grassland" areas for conservation. The regal fritillary butterfly may be acting as an indicator species; warning that a valuable habitat is critically endangered and that present open area land management practices in the Northeast should be reevaluated.

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