

THE PHYLOGENETICS AND BIOCHEMISTRY OF HOST-PLANT SPECIALIZATION IN MELITAEINE BUTTERFLIES (LEPIDOPTERA: NYMPHALIDAE)

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Abstract.—Butterflies in the tribe Melitaeini (Lepidoptera: Nymphalidae) are known to utilize host plants belonging to 16 families, although most host-plant records are from four families. Of the 16 host-plant families, 12 produce secondary plant metabolites called iridoids. Earlier studies have shown that larvae of several melitaeine species use iridoids as feeding stimulants and sequester these compounds for larval defense. I investigate the evolutionary history of host-plant use in the tribe Melitaeini by testing a recent phylogenetic hypothesis of 65 species representing the four major species groups of the tribe. By simple character optimization of host-plant families and presence/absence of iridoids in the host plants, I find that plant chemistry is a more conservative trait than plant taxonomy. The ancestral host plant(s) of the entire tribe most likely contained iridoids and were likely to be in the plant family Plantaginaceae. A major host shift from plants containing iridoids to plants not containing iridoids has happened three times independently. The results show that the evolution of host-plant use in melitaeines has been (and still is) a dynamic process when considering plant taxonomy, but is relatively stable when considering host-plant chemistry.

Key words.—Asteridae, host-plant use, iridoid glycosides, Lepidoptera, Melitaeini.

Received March 13, 2000. Accepted October 18, 2000.

Phytophagy and the large diversity of species in insects are closely linked phenomena that researchers have been attempting to explain for the past several decades. The two clear patterns are that phytophagous clades are more species rich than their nonphytophagous sister groups (Mitter et al. 1988) and most phytophagous insects are highly specialized on a few or even only one species of host plant (Dethier 1954; Ehrlich and Raven 1964; Futuyma 1983). To explain the former pattern, one must first understand the latter pattern. One way to achieve this is to attempt to reconstruct the evolutionary history of host-plant use in a group of related insects. With the almost exponential increase in the availability of phylogenetic hypotheses due to advances in molecular systematics, the potential to rigorously test the different hypotheses of insect–host plant relationships is today great.

There are several hypotheses that aim to explain the high specialization observed in phytophagous insects, such as through coevolution of insects and their host plants (Ehrlich and Raven 1964) or through insects simply colonizing plants without affecting their evolution (Jermy 1984, 1993). A common theme in most hypotheses concerning insect–host plant associations is that plant secondary chemistry is the major constraining force behind the patterns we see today in the use of host plants by insects (Brower and Brower 1964; Feeny 1991; Futuyma 1991; Becerra 1997; Nylin and Janz 1999).

The influence of host-plant chemistry on the evolution of host-plant use has been shown in a study of the leaf beetle genus *Blepharida* (Becerra 1997). Related species of beetle were more likely to be found on chemically similar plant species, regardless of the phylogenetic relationship of the plants. Other studies have inferred that ancestral host plants are more likely to be recolonized than totally new host plants (Futuyma and McCafferty 1990; Funk et al. 1995). Most published studies have considered leaf beetles in the family Chrysomelidae and single studies have usually concentrated

on the evolution of host-plant use in one genus (e.g., Futuyma and McCafferty 1990; Funk et al. 1995; Dobler et al. 1996; Becerra 1997; Mardulyn et al. 1997; Köpf et al. 1998). In this paper, I infer the evolutionary history of host-plant use in a tribe of butterflies using methods similar to those used in the previously cited studies.

Members of the order Lepidoptera are almost entirely phytophagous (Powell et al. 1999). Within Lepidoptera, the butterflies (Papilionoidea) have received most of the attention of researchers interested in insect–host plant relations (Brower and Brower 1964; Ehrlich and Raven 1964; Benson et al. 1975; Miller 1987; Brown et al. 1991; Feeny 1991; Janz and Nylin 1998; Nylin and Janz 1999; Powell et al. 1999). The general patterns of host-plant use in butterflies reported by Ehrlich and Raven (1964)—that related species of butterflies tend to use related species of host plants—have been confirmed in a more phylogenetically rigorous study by Janz and Nylin (1998). The pattern is strengthened by the inference that ancestral host plants are more easily recolonized than completely novel host plants are colonized, as with the beetles (Futuyma and McCafferty 1990; Funk et al. 1995). Janz and Nylin (1998) also reported that butterflies use mainly plants in families that diversified before butterflies themselves diversified.

The importance of certain chemicals to host use has been established for several groups of butterflies that feed on a restricted set of plant species. In many cases, specialization of butterfly species on to plant species has led to the larvae utilizing the defense chemicals of the plants in their own defense (Brower and Brower 1964). Often larvae sequester only one class of secondary plant metabolites, even though the plants usually contain other classes of secondary metabolites. Well known examples include species in the subfamily Danainae (Nymphalidae), which specialize on plants in the family Apocynaceae (which includes Asclepiadiaceae in the present classification of plants; Angiosperm Phylogeny Group 1998 [henceforth APG 1998]) that contain cardenol-

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ides (Brower 1984), and many species of *Papilio* (Papilionidae), which are found only on plants in the families Rutaceae and Apiaceae that contain furanocoumarins (Berenbaum 1983; Feeny 1991).

While the associations of groups of butterfly species with groups of plant species has been widely acknowledged, rigorous phylogenetic studies looking at the evolution of these associations are lacking. Without the benefit of phylogenetic hypotheses, earlier studies came to conclusions that are perhaps too simplified. Recent studies using cladograms have shown that the evolution of host-plant use is a dynamic process, rather than a steady progression from less specialized to more specialized (Miller 1987; Radtkey and Singer 1995; Brower 1997; Janz et al. 2001).

Here, I investigate the pattern of host-plant use of butterflies in the tribe Melitaeini (sensu Harvey 1991) using simple character optimization onto a cladogram. Melitaeini belongs to the family Nymphalidae and comprises about 250 species that are distributed throughout the Palaearctic, Nearctic, and Neotropical regions (Higgins 1981). The relationships of genera and species within them have been investigated in a recent molecular phylogeny of the tribe (Wahlberg and Zimmermann 2000; Fig. 1).

The host plants of melitaeine butterflies belong largely to the subclass Asteridae sensu lato, with the plant families Acanthaceae, Asteraceae, Orobanchaceae, and Plantaginaceae being the most important (Ackery 1988; Fig. 2). Most of the major host-plant families utilized by melitaeine butterflies produce secondary chemicals known as iridoids (Jensen et al. 1975). Iridoids are divided into two major groups: iridoid glycosides and seco-iridoids. Both are known to be extremely bitter and can have emetic effects on vertebrates (Bowers 1980; Seigler 1998). The distribution of these two groups of chemicals does not overlap in the plant families used by melitaeines (Fig. 2). Bowers (1983) has shown that iridoid glycosides are necessary feeding stimulants for larvae of Nearctic species in the genus *Euphydryas* and that the larvae are able to sequester these compounds for use in their own chemical defense. *Euphydryas* larvae are considered aposomatic and they are unpalatable to birds (Bowers 1980, 1981). Larvae of other melitaeine genera are also known to sequester iridoid glycosides (L'Empereur and Stermitz 1990; Mead et al. 1993; Stermitz et al. 1994; Lei and Camara 1999). One of the major host-plant families, Asteraceae, lacks iridoids completely (Jensen et al. 1975; El-Naggar and Beal 1980; Boros and Stermitz 1990; Jensen 1991), although closely related plant families contain them (Fig. 2).

In this paper I investigate how important host plant taxonomy is to their utilization by a species in the butterfly tribe Melitaeini and to what extent plant secondary compounds are involved in the specialization of melitaeine species.

MATERIALS AND METHODS

The phylogenetic hypothesis for the melitaeines used in this paper is taken from a cladistic analysis of mitochondrial DNA (mtDNA) sequences by Wahlberg and Zimmermann (2000), with additional information on the position of *Phyciodes vesta*. Wahlberg and Zimmermann (2000) sequenced 1422 bp from the cytochrome oxidase I (COI) gene and 536

bp from the 16S ribosomal gene from 77 species of melitaeines, which represent all major groups in the tribe. The position of *P. vesta* in the cladogram was inferred from a cladistic analysis of the COI dataset of Wahlberg and Zimmermann (2000), which included the *P. vesta* sequence (N. Wahlberg, unpubl. data). The COI gene of *P. vesta* was sequenced (1422 bp) according to the protocol given in Wahlberg and Zimmermann (2000). The position of *P. vesta* had strong jackknife support (80% on the branch leading to clade 4 in Fig. 1). Figure 1 gives the phylogenetic hypothesis used in this paper. Species for which there was no information on host-plant use have been pruned from figure 4 in Wahlberg and Zimmermann (2000).

I was able to find host-plant records for 65 species included in the cladogram (Appendix). Data on host-plant use were mainly taken from the literature (White and Singer 1974; Williams et al. 1984; Luckens 1985; Scott 1986, 1994, 1998; DeVries 1987; Weidemann 1988; Thomas et al. 1990; Ebert 1991; Dyer and Floyd 1993; Hesselbarth et al. 1995; Kuussaari et al. 1995; Denno and Benrey 1997; Klemetti and Wahlberg 1997; Tolman 1997; Wahlberg 1997, 1998; Feldman and Haber 1998), but also some unpublished records were used (N. Wahlberg, pers. obs.; M. Singer, pers. comm. 1999; S. van Nouhuys, pers. comm. 2000).

The classification of flowering plants has recently undergone a radical change with the advent of molecular systematics (Chase et al. 1993; APG 1998). I used the most recent classification of Asteridae in this work (Olmstead et al. 1993, 2001; Olmstead and Reeves 1995; APG 1998; Fig. 2). This classification departs from the traditional classification in several respects. Most importantly for this paper, Scrophulariaceae has been found to be polyphyletic and species belonging to the traditional Callitrichaceae, Globulariaceae, Plantaginaceae, and Scrophulariaceae have been placed in Plantaginaceae (e.g., *Veronica*, *Plantago*), Orobanchaceae (e.g., *Melampyrum*, *Castilleja*, *Pedicularis*), and Scrophulariaceae (e.g., *Verbascum*, *Buddleja*; Judd et al. 1999; Olmstead et al. 2001). Olmstead et al. (2001) refer to the family Plantaginaceae as the clade Veronicaceae.

Melitaeines can be considered to be specialized, using one or two host plant species at the population level (e.g. White and Singer 1974), however, a species as a whole may be associated with up to 20 plant species in up to six plant families. It is thus important to choose the appropriate hierarchical level for the host character (Janz 1999). In this case, the level of plant species or even genus is probably too labile for useful analysis at the level of butterfly species (Radtkey and Singer 1995). I therefore chose the level of plant family as my host-plant characters. For each butterfly species I coded the use of host-plant family as a binary character. This overcomes the problems associated with attempting to optimize polymorphisms, but may produce inaccurate reconstructions of internal branches as having no host-plant associations at all (Janz 1999). In such cases, I treated the host association as equivocal. All character optimizations were performed using the program MacClade 3.05 (Maddison and Maddison 1992).

I investigated the role of iridoids in the evolution of host-plant associations of the melitaeines by coding the presence of iridoid glycosides and seco-iridoids as separate binary

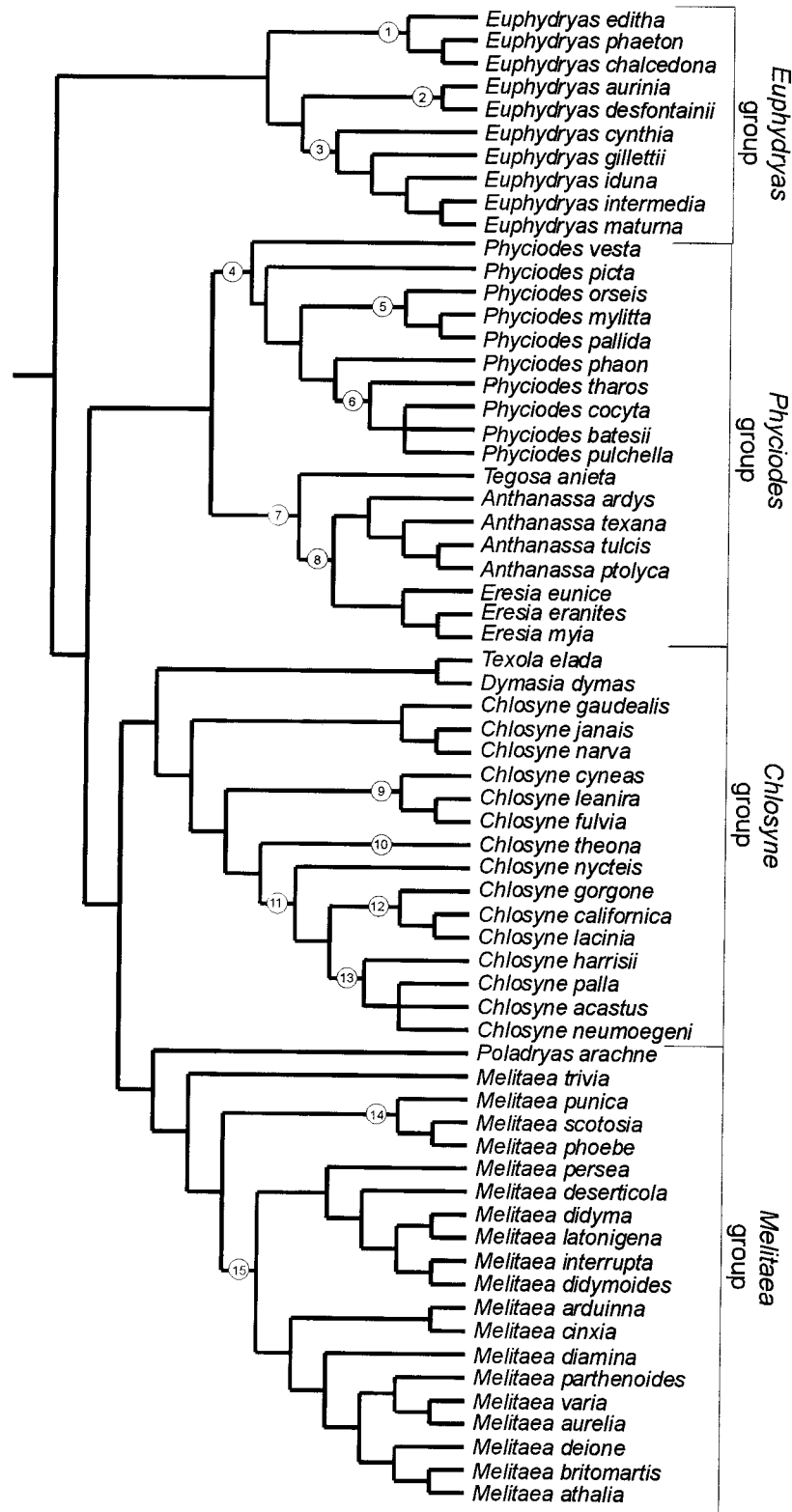


FIG. 1. A molecular phylogeny of the tribe Melitaeini based on 1422-bp COI and 536-bp 16S sequences (Wahlberg and Zimmermann 2000). The numbered clades are discussed in the text.

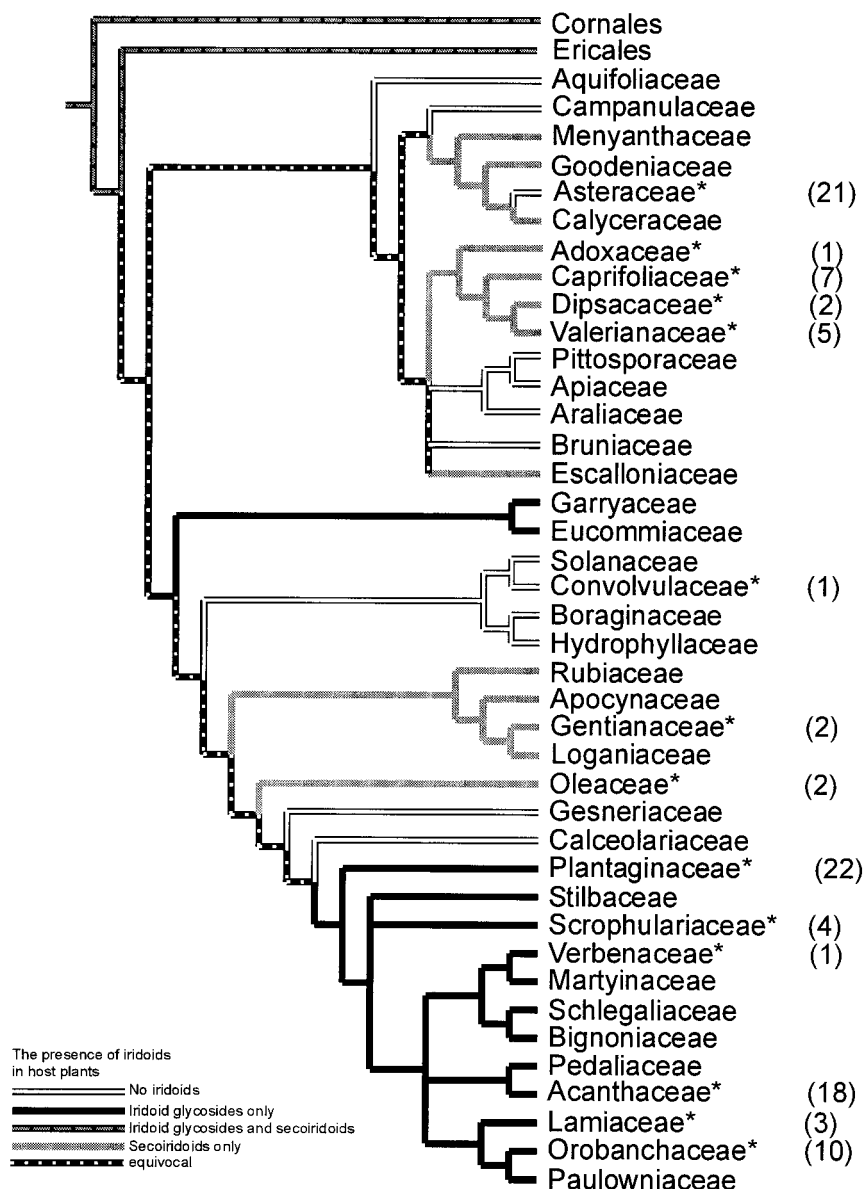


FIG. 2. The most recent molecular phylogeny of the subclass Asteridae based on Olmstead et al. (1993), Olmstead and Reeves (1995), Angiosperm Phylogeny Group (1998), and Olmstead et al. (2001). The presence of iridoid glycosides and seco-iridoids has been mapped onto the phylogeny for illustrative purposes only and is not meant to be a rigorous study of the evolution of iridoids in Asteridae. Plant families marked with an asterisk are used by species of Melitaeini. Numbers in parentheses show the number of species used in this study that use plants in the family to the left of the number.

characters. Most host-plant species used by melitaeine species have not been bioassayed for iridoids, but the compounds are ubiquitous in the plant genera to which the host plants belong in the plant families Adoxaceae, Caprifoliaceae, Dipsacaceae, Gentianaceae, Oleaceae, Orobanchaceae, Plantaginaceae, Scrophulariaceae, Valerianaceae, and Verbenaceae (Jensen et al. 1975; El-Naggar and Beal 1980; Boros and Stermitz 1990; Jensen 1991; S. R. Jensen, pers. comm. 2000). The presence of iridoids in Acanthaceae is patchy (Jensen et al. 1988) and apparently they occur mainly in basal lineages of the plant family (cf. Jensen et al. 1988 and McDade and Moody 1999). Iridoids are not known to occur in the acanth genera used by melitaeines (S. R. Jensen, pers. comm. 2000).

Iridoids are also not known to occur in Amaranthaceae, Asteraceae, and Convolvulaceae (Hegnauer 1964; Jensen 1991; Fig. 2).

In the above analyses, gains of plant families or plants containing iridoids are considered to be equal to losses. However, a loss of a host-plant family also may arise from the loss of the plant family from the habitat and as such does not entail a genetic change in the insect itself. The gain of a new plant family in the repertoire of an insect, however, may necessitate a genetic change in the oviposition behavior of adult females or in the ability of the larvae to utilize the new resource. To take this asymmetry into account, I investigated the effects of making a gain more costly than a loss.

Because the costs of gaining new host plant families or colonizing plants with different biochemistry are unknown, I have explored different scenarios by making gains 1.5, 2.0, 3.0, and 4.0 times as costly as losses.

To investigate how phylogenetically conservative host-plant family use and host plant chemistry are, I used a modified version of the PTP test (Faith and Cranston 1991) on single characters rather than the entire dataset. I randomly reshuffled each character separately among the species 300 times and compared the number of steps required to explain each random reshuffle to the observed number of steps for that character. This gives a probability that the observed pattern does not differ from random that is $(n + 1)/300$, where n is the number of replications with less than or equal number of steps to the observed number of steps. This value is thus minimally 1/300, or 0.003. The reshuffling of character states was done with the equally weighted dataset in the computer program MacClade 3.05 (Maddison and Maddison 1992).

RESULTS

Tracing plant families on to the melitaeine phylogeny indicates that host-plant utilization is dynamic at the level of host-plant family (Fig. 3). The most parsimonious solution indicates a total of 32 unambiguous independent colonizations of 12 plant families, while losses were recorded unambiguously only twice in two families (Table 1). The 10 ambiguities can be reconstructed in two ways, a character may be optimized so that gains are more likely to be homologies that are subsequently lost (known as fast or ACCTRAN optimization) or they may be optimized so that gains are more likely to be homoplasious (i.e., independent gains—slow or DELTRAN optimization; see Kitching et al. 1998). The five most commonly used host plant families showed ambiguities when optimized on to the cladogram (Table 1). This may come about through multiple losses and recolonizations of these host-plant families, which tends to blur phylogenetic information.

I was unable to recover the ancestral host-plant family of the butterfly tribe, or even for the four species groups, unambiguously when gains and losses were weighted equally (Fig. 3). The *Euphydryas* group has the broadest host-plant family repertoire of Melitaeini (Fig. 3), utilizing, on average, host-plant species in three families. The butterfly species in the other three species groups are largely found on plants in only one family. The ancestor of the *Euphydryas* group probably specialized on plants in Plantaginaceae and Caprifoliaceae. The use of host-plant families by *Euphydryas* species is very dynamic, with repeated independent colonizations inferred for several plant families (Orobanchaceae, Oleaceae, and Valerianaceae). It is interesting to note that the host-plant family utilization of the Nearctic *E. gillettii* has converged to the other Nearctic species in the *E. editha* clade (number 1 in Fig. 1; Fig. 3). The use of plants in Plantaginaceae is inferred to have been lost twice and the use of plants in Caprifoliaceae has been lost three times.

The ancestral host-plant family of the *Phyciodes* group was not recovered (Fig. 3). However, the major families in this species group are Acanthaceae and Asteraceae. The *Phyciodes* group is composed of two biogeographical clades (Wahl-

berg and Zimmermann 2000), species in the *Phyciodes* clade (number 4 in Fig. 1) are found in the Nearctic and species in the *Tegosa-Anthanassa-Eresia* clade (number 7 in Fig. 1) are found in the Neotropics. The Nearctic species are mainly specialized on plants in the family Asteraceae. Species of *Tegosa* use only host plants in Asteraceae (DeVries 1987; Brown 1992). The *Anthanassa-Eresia* clade (number 8 in Fig. 1) contains about 100 species and all species with known host plants use species in Acanthaceae. Within the *Phyciodes* s.s. clade, three species depart from the general pattern of utilization of host plants only in Asteraceae. *Phyciodes picta* has also colonized Convolvulaceae, an association that is recent because the plant species was introduced from Europe to North America by humans (Scott 1994). *Phyciodes vesta* and *P. phaon* are found on plant species in Acanthaceae and the latter species has also colonized Verbenaceae. Neither species are known to use plants in Asteraceae.

Acanthaceae is also the major host-plant family of the basal species in the *Chlosyne* group and is thus inferred to be the ancestral host-plant family of this group (Fig. 3). Going up the clade, there is an inferred colonization of Orobanchaceae with an associated loss of Acanthaceae. The most derived clade in the *Chlosyne* group has colonized Asteraceae and simultaneously lost the use of Orobanchaceae. In addition to these general patterns, there are single colonizations of plant families by single species: Asteraceae by *Texola elada*, Amaranthaceae by *Chlosyne narva*, and Scrophulariaceae by *Chlosyne theona*.

The ancestral host-plant family of the *Melitaea* group is not as clear (Fig. 3). The most basal species, *Poladryas arachne* (the only Nearctic representative of this otherwise Palearctic species group), utilizes plants in the family Plantaginaceae, which is an ancestral host plant family in the large *Melitaea* clade number 15 (Figs. 1, 3). Between the *Poladryas* branch and the large *Melitaea* clade are two branches that have colonized different host-plant families. *Melitaea trivialis* is found only on *Verbascum* species (Scrophulariaceae; Tolman 1997) and the *M. phoebe* clade (number 14 in Fig. 1) is only found on plants in Asteraceae. Within the larger *Melitaea* clade, there have been several colonizations of new host-plant families by single species: a widening of host range by *M. didyma* onto Lamiaceae and Scrophulariaceae, *M. varia* onto Gentianaceae and *M. athalia* onto Orobanchaceae, and host shifts by *M. diamina* onto Valerianaceae and *M. arduinna* onto Asteraceae.

The five most commonly used host-plant families (Acanthaceae, Asteraceae, Caprifoliaceae, Orobanchaceae, and Plantaginaceae) have a distinct phylogenetic signal, despite multiple colonizations and losses. All five families require fewer steps when optimized onto the cladogram than the same data randomized over the taxa ($P = 0.003$ in all cases). The implication is that once a plant family is colonized, descendant species tend to continue using host plants in that family. All other plant families are inferred to have been colonized by single species (i.e., are autapomorphic) or have been colonized a single time by a small clade of species.

When asymmetrical costs are invoked, the number of independent colonizations of the five most commonly used host plants decreases rapidly (Table 2). Three plant families Acanthaceae, Caprifoliaceae, and Plantaginaceae, are optimized

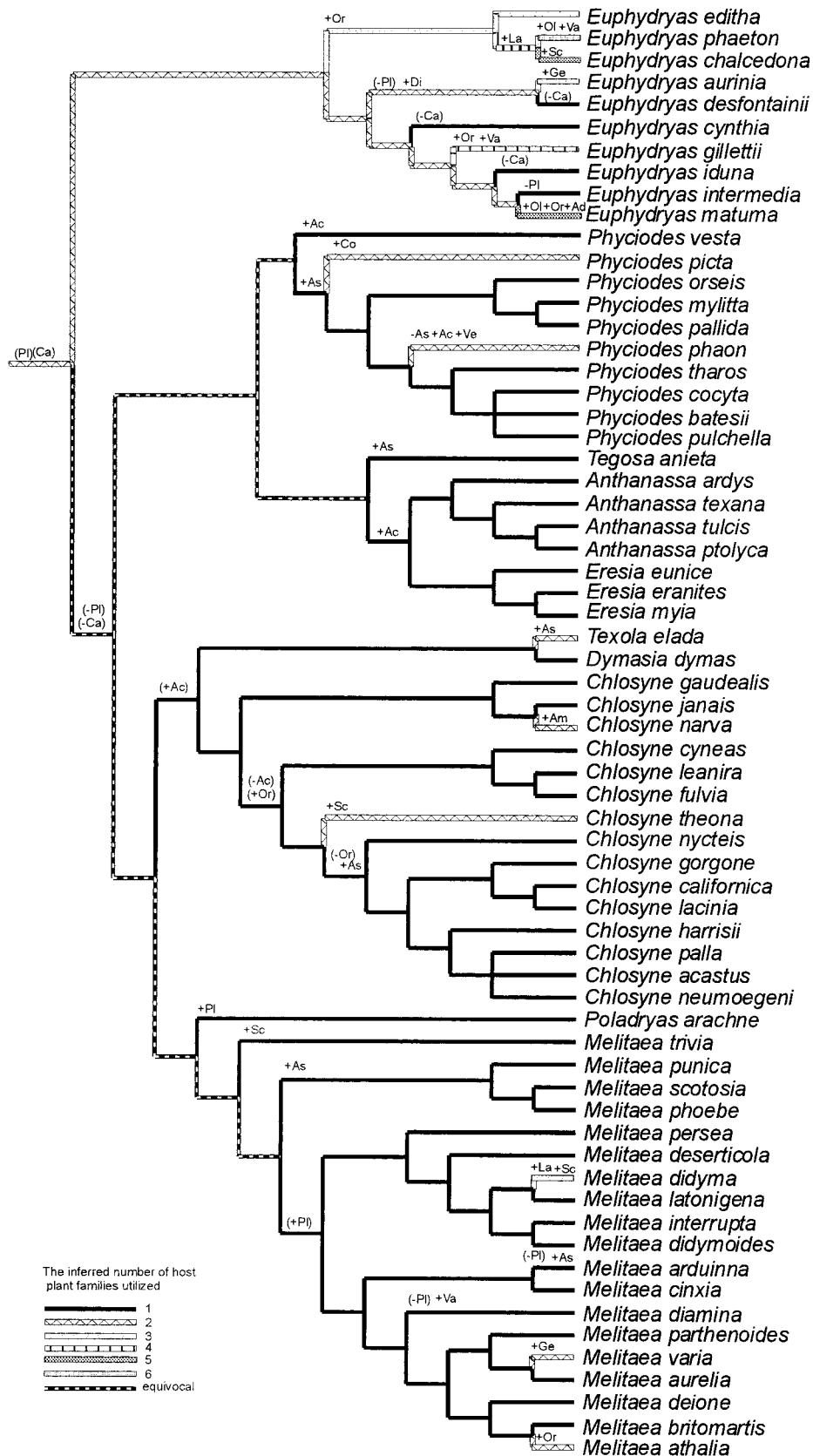


FIG. 3. Optimization of host-plant families used by melitaeine butterflies. Gains (+) and losses (-) are shown above the relevant branches. The plant families have been optimized as independent binary characters. Abbreviations for plant families are as follows: Ac, Acanthaceae; Ad, Adoxaceae; Am, Amaranthaceae; As, Asteraceae; Ca, Caprifoliaceae; Co, Convolvulaceae; Di, Dipsacaceae; Ge, Gentianaceae; La, Lamiaceae; Ol, Oleaceae; Or, Orobanchaceae; Pl, Plantaginaceae; Sc, Scrophulariaceae; Va, Valerianaceae; Ve, Verbenaceae. Parentheses around the plant family abbreviations denote ambiguities resolved using fast optimization. Branch shading indicates the inferred number of plant families used by ancestral populations.

TABLE 1. A summary of the colonizations and losses of host-plant family use according to three optimization procedures. Unam, unambiguous changes in characters; fast, ambiguities in character optimization reconstructed so that colonizations minimized and losses maximized; and slow, ambiguities in character optimization reconstructed so that colonizations maximized and losses minimized.

Host-plant family	Colonizations			Losses		
	Unam	Fast	Slow	Unam	Fast	Slow
Acanthaceae	3	4	5	0	1	0
Adoxaceae	1	1	1	0	0	0
Amaranthaceae	1	1	1	0	0	0
Asteraceae	6	6	6	1	1	1
Caprifoliaceae	0	0	4	0	4	0
Convolvulaceae	1	1	1	0	0	0
Dipsacaceae	1	1	1	0	0	0
Gentianaceae	2	2	2	0	0	0
Lamiaceae	2	2	2	0	0	0
Oleaceae	2	2	2	0	0	0
Orobanchaceae	4	5	6	0	1	0
Plantaginaceae	1	2	6	1	5	1
Scrophulariaceae	4	4	4	0	0	0
Valerianaceae	3	3	3	0	0	0
Verbenaceae	1	1	1	0	0	0
Total	32	35	45	2	12	2

down to the root of the cladogram when gains are made 1.5 times more costly than losses. Making gains twice as costly as losses places Orobanchaceae unambiguously at the root and Asteraceae at the root when fast optimization is used. Increasing the cost asymmetry further places all five commonly used host-plant families at the root.

The presence of iridoid glycosides in the ancestral host plants of the melitaeines was not recovered unambiguously, although the ancestral host-plant family of the entire butterfly tribe is inferred to have contained the chemicals when fast optimization is used. According to the hypothesis presented in Figure 4, there have been nine independent losses of the character state (Table 3). Six of these losses correspond to an inferred colonization of plants in Asteraceae or Acanthaceae. Three of the losses of iridoid glycosides are by species that have shifted to host plants containing only seco-iridoids.

There has been one inferred recolonization of plants containing iridoid glycosides in the *Phyciodes* group, by a single species, *P. phaon*.

Plants containing seco-iridoids have been used mainly by species in the *Euphydryas* group, where at least one ancestral host plant species is inferred to have contained the compounds (Fig. 4). Within the *Euphydryas* group, there have been two independent losses of use of plants containing seco-iridoids, once by the alpine *E. cynthia* and once by the arctic *E. iduna*. In addition to these, there appear to have been two independent colonizations of plants containing seco-iridoids in the *Melitaea* group. The colonization by *M. diamina* onto Valerianaceae is associated with a host shift, whereas *M. varia* has widened its host range by colonizing Gentianaceae.

The phylogenetic signal of both types of compounds is strong ($P = 0.003$ for both compounds). In the case of iridoid glycosides, ancestral populations that have lost the utilization of plants containing these chemicals tend to remain on plants that do not contain iridoid glycosides. The colonization of plants containing seco-iridoids is also phylogenetically conservative. Invoking asymmetrical costs by making the gain of an iridoid-containing plant family more costly than the loss of such a host-plant family removes the ambiguity apparent in the equally weighted scheme.

DISCUSSION

The results I have presented in this paper are highly dependent on the phylogenetic hypothesis available. However, my results are fairly robust when one considers the branch support values given in Wahlberg and Zimmermann (2000). Collapsing branches with low support (which correspond to the branches connecting the four species groups) does not affect the general results of this paper. The cladogram that I have used in this paper contains only about a quarter of the known species of Melitaeini, and the four species groups are differentially represented. The *Euphydryas* group has only two species missing, both of which are closely related to *E. aurinia* (Zimmermann et al. 2000). The host plants of these two species are unknown, but given the specialization of *E.*

TABLE 2. The effects of increasingly asymmetrical costs in host-plant family optimizations. Root indicates that the plant family was optimized down to the root of the cladogram, that is, it is inferred to be the ancestral host-plant family of the entire butterfly tribe. Parentheses indicate that an ambiguity was resolved using fast optimization.

Host-plant family	Cost of gains to losses				
	1.0	1.5	2.0	3.0	4.0
Acanthaceae	4–5 gains 0 losses	root 1 gain 5 losses	root 0–1 gain 5–7 losses	root 0 gains 7 losses	root 0 gains 7 losses
Asteraceae	6 gains 1 loss	6 gains 1 loss	(root) 0–6 gains 1–13 losses	root 0 gains 13 losses	root 0 gains 13 losses
Caprifoliaceae	(root) 0–4 gains 0–4 losses	root 0 gains 4 losses	root 0 gains 4 losses	root 0 gains 4 losses	root 0 gains 4 losses
Orobanchaceae	5–6 gains 0–1 losses	5 gains 1 loss	root 2 gains 6 losses	root 1–2 gains 6–10 losses	root 1 gain 10 losses
Plantaginaceae	(root) 2–6 gains 1–5 losses	root 0–2 gains 4–8 losses	root 0 gains 8 losses	root 0 gains 8 losses	root 0 gains 8 losses



FIG. 4. Optimization of presence of two classes of iridoids in the host plants of melitaeine species. Fast optimization used. The two classes have been optimized independently but are shown here together to save space.

aurinia and *E. desfontainii* on plants containing only secoiridoids, it is quite likely that the missing species are also similarly specialized.

The *Phyciodes* group is represented by 18 species of an estimated 130 species (Wahlberg and Zimmermann 2000).

Only one species (host plant unknown) is missing from clade 4 (Fig. 1), while the rest of the more than 100 species putatively belong to clade 7 (Fig. 1). The host plants of most species are unknown, but those species with known host plants feed on plants in Acanthaceae (six species whose phy-

TABLE 3. A summary of the colonizations and losses of host plants containing iridoid glycosides and/or seco-iridoids according to three optimization procedures. Column subheadings as in Table 1.

Type of iridoids	Colonizations			Losses		
	Unam	Fast	Slow	Unam	Fast	Slow
Iridoid glycosides	1	1	6	4	9	4
Seco-iridoids	3	3	3	2	2	2

logenetic position is unknown; DeVries 1987; Brown 1992), with the exception of *Tegosa* species, which feed on Asteraceae. The evolutionary history of this large group deserves closer attention because it appears the ancestor of the group has colonized the Neotropics from the Nearctic region (Wahlberg and Zimmermann 2000).

The *Chlosyne* group is missing 13 species, of which six putatively belong to clade 11 (Fig. 1) and the rest are likely to be more basal. The former species all use plants in Asteraceae (Scott 1986), conforming to the patterns shown here, while all of the latter species have unknown host associations. The *Melitaea* group is missing 35 species, for which the host-plant associations are largely unknown. One species (*M. asteria*) belonging to clade 15 (Fig. 1) is known to use a species of *Plantago* (Plantaginaceae; Tolman 1997).

In summary, the available phylogenetic hypothesis for the tribe Melitaeini gives a sufficiently good representation of the evolutionary history of host-plant use at the level of plant families. The results of my study indicate that the evolution of host-plant utilization is a fairly dynamic trait in the tribe Melitaeini and that the presence of iridoid glycosides in the host plants has had a major influence on the evolutionary history of host-plant use. Because iridoids are mainly found in one, albeit large, clade of plants (Jensen et al. 1975; Fig. 2), the pattern of host-plant use in Melitaeini shows the taxonomic conservatism that has been described for many other phytophagous insect groups (Ehrlich and Raven 1964; Futuyma 1983; Strong et al. 1984; Mitter and Farrell 1991; Janz and Nylin 1998). Even the most prominent colonizations of plants that do not contain iridoids are restricted to two plant families (Acanthaceae and Asteraceae), which belong to the same clade as the iridoid-containing plants. There are in fact only two known associations of melitaeine species with plants outside Asteridae (sensu APG 1998). These are *C. narva* on Amaranthaceae (DeVries 1987) and *Eresia alsina* on Urticaceae (as *Eresia eutropia* in Young 1973; see DeVries 1987). The latter species was not available when the cladogram was generated and is thus not shown in the figures.

Although my results are somewhat ambiguous at the deeper levels of the cladogram in relation to the actual plant family used by ancestral species, it appears clear that the plant family (or families) contained iridoid glycosides. Plantaginaceae is the only plant family that contains iridoid glycosides that is placed consistently at the root when imposing increased costs to gaining plant families. This suggests that the ancestral population of the entire butterfly tribe used Plantaginaceae and that the utilization of this plant family has been lost independently by the *Phycodes* group and the *Chlosyne* group.

Because the presence of iridoid glycosides is optimized

down to the root, one can hypothesize that iridoid glycosides may be important to species in the sister group of the melitaeines. The putative sister clade of the Melitaeini is the tribe Kallimini (Harvey 1991; Brower 2000). Species in the Kallimini (such as those in the genera *Junonia*, *Hypolimnas*, *Anartia*, and *Siproeta*) are known to specialize on plants in Acanthaceae and Plantaginaceae (Scott 1986; DeVries 1987), lending support to the suggestion that at least the latter plant family was used by the ancestral population of Melitaeini. At least one kallimine species (*Junonia coenia*) is known to specialize on iridoid glycosides (Bowers 1984). Thus, it appears that the association with plants containing iridoid glycosides is more ancient than the melitaeine tribe itself.

Plants containing seco-iridoids are generally not used by melitaeines. The only clade in which seco-iridoid-containing plants are utilized is that of the *Euphydryas* group. Those species that do utilize plants containing seco-iridoids use plants that also contain iridoid glycosides (see Fig. 2). The exception is the *Euphydryas aurinia* clade (number 2 in Fig. 1), which mainly utilizes plants in Dipsacaceae containing only seco-iridoids. *Euphydryas desfontainii* and the majority of *E. aurinia* populations are known to only use plants that belong to Dipsacaceae. A few populations of *E. aurinia* are specialized on species in Caprifoliaceae or Gentianaceae, which also contain seco-iridoids.

Melitaeine species using plants containing iridoid glycosides are known to utilize these chemicals in larval defense and as larval phagostimulants. In the *Euphydryas* group, *E. editha*, *E. phaeton*, *E. chalcedona*, *E. cynthia*, and *E. gillettii* have been shown to sequester iridoid glycosides as larvae (Bowers 1983; Bowers and Puttick 1986; Stermitz et al. 1986; Franke et al. 1987; Williams and Bowers 1987; Gardner and Stermitz 1988). In the *Chlosyne* group, *C. leanira* and *C. theona* are known to sequester the compounds (Mead et al. 1993; Stermitz et al. 1994), and in the *Melitaea* group, at least *Poladryas arachne* and *Melitaea cinxia* have this ability (L'Empereur and Stermitz 1990; Lei and Camara 1999). An interesting point is that all the studies mentioned above have found only five of more than 500 iridoid glycosides reported (Seigler 1998) being sequestered. These are aucubin, catalpol, macfadienoside, methyl shanzhiside, and 6- β -hydroxyipolamide. The first two iridoid glycosides are sequestered by almost all species studied, whereas the latter three have been found in only single species. *Euphydryas gillettii* larvae are unable to sequester seco-iridoids from their host plant *Lonicera involucrata* (Caprifoliaceae; Bowers and Williams 1995), although they are able to sequester iridoid glycosides from rarely used host plants in Plantaginaceae. Whether the two species of *Euphydryas* that specialize on plants containing only seco-iridoids are able to sequester these compounds is unknown.

The evolutionary history of host-plant use in the melitaeines is somewhat linked to the biogeography of the group. The Nearctic species in the *Euphydryas* group are able to exploit a wide range of plants containing both iridoids, but most populations specialize on Plantaginaceae and Orobanchaceae (Scott 1986; Thomas et al. 1990; Radtkey and Singer 1995), which contain only iridoid glycosides. It may be that there is a cost associated with using plants containing seco-iridoids. The Palaearctic clade split further into a southern

and a northern clade, the *E. aurinia* clade (number 2 in Fig. 1) and *E. cynthia* clades (number 3), respectively (Zimmermann et al. 2000). The southern clade became specialized on Dipsacaceae, which contain only seco-iridoids. Species in the northern clade utilize host plants in a variety of plant families, although individual species may be restricted to one plant family. The loss of Caprifoliaceae in *E. cynthia* and *E. iduna* may just be due to the habitat of these two species. The alpine (*E. cynthia*) and arctic (*E. iduna*) areas are not amenable to growth by species of Caprifoliaceae. *Euphydryas gillettii* has recolonized the Nearctic area and seems to have converged in host-plant use to the other Nearctic species. It differs in one respect, its main host plant is in Caprifoliaceae. The convergence is likely to be a result of the flora being more similar within the Nearctic compared to between the Nearctic and Palearctic.

Nearctic species in the *Phyciodes* and *Chlosyne* groups tend to be associated with Asteraceae, whereas their Neotropical counterparts are mostly associated with Acanthaceae. The ancestral host plant family of both species groups is ambiguous when gains and losses are equally weighted. Acanthaceae is unambiguously ancestral in both species groups when imposing increased costs to gaining host plant families, whereas the ancestral use of Asteraceae remains ambiguous until the highly asymmetric costs of 1:3 (losses:gains; Table 2). According to this scenario, there have been two independent host shifts in the *Phyciodes* group from Acanthaceae to Asteraceae. In the *Chlosyne* group, there have been two host shifts, one from Acanthaceae to Orobanchaceae and then from Orobanchaceae to Asteraceae. The association of biogeography with host-plant family is an intriguing mystery that requires a more complete phylogenetical hypothesis for both species groups.

As far as is known, Acanthaceae is not used at all by Palearctic species of Melitaeini, which are mainly found on Plantaginaceae. This can possibly be attributed to the fact that Plantaginaceae are largely temperate plants, whereas the Acanthaceae are largely tropical plants. The Old World melitaeines have not colonized the tropical regions and therefore have not come into contact with plants belonging to Acanthaceae with sufficient frequency for these plants to be incorporated into their diets. Old World Acanthaceae species may also be unsuitable to melitaeines, as larvae of *Anthanassa ardys* and *Anthanassa tucis* are unable to complete development on an African species of Acanthaceae introduced to Costa Rica (Feldman and Haber 1998).

Utilization of plants in Asteraceae by melitaeines is fairly limited. Within the *Phyciodes s.s.* clade (number 4 in Fig. 1), species in the *Phyciodes mylitta* clade (number 5 in Fig. 1) are known to feed only on species in the Asteraceae tribe Cardueae and species in the *Phyciodes tharos* clade (number 6 in Fig. 1) and *P. picta* feed only on species in the tribe Astereae. *Tegosa anieta* and *T. claudina* are known to feed on *Mikania* sp. (DeVries 1987; Freitas 1991), which belong to the tribe Eupatorieae. In the *Chlosyne* group, *C. nycteis* feeds on a variety of Asteraceae in the tribes Astereae and Heliantheae. The two more derived clades are found on a subset of these host-plant tribes, the *Chlosyne gorgone* clade (number 12 in Fig. 1) feeds on Heliantheae and the *Chlosyne palla* clade (number 13) is found on plants in the Astereae.

All species in the *Melitaea* group that feed on plants in Asteraceae are restricted to the tribe Cardueae. What feature of the particular tribes within Asteraceae make them amenable to colonization by butterflies that otherwise show high fidelity to plants that are chemically different to Asteraceae? At the moment there is no definitive answer to this question. There may be some other group of chemicals in common to the plant families used by melitaeines, although I have been unable to find a clear-cut group (see Seigler 1998).

In this study I have shown that host-plant chemistry is a more conservative feature than host-plant taxonomy in the evolutionary history of host-plant use in a tribe of butterflies. Host-plant taxonomy is important at a lower level in single clades within the tribe, which can be seen in the strong phylogenetic signals given by the five most commonly used host-plant families. I tentatively conclude that the most likely ancestral host plant(s) contained iridoid glycosides and belonged to the plant family Plantaginaceae. This conclusion can be tested with a good phylogenetic hypothesis for the subfamily Nymphalinae (to which Melitaeini belongs), which would resolve the extent of the association with plants containing iridoid glycosides and plants in the family Plantaginaceae. My study has placed the patterns of host-plant use in the tribe Melitaeini in a historical perspective. This perspective will help to identify subjects for future study, such as why have noniridoid plants been colonized independently several times.

ACKNOWLEDGMENTS

I am grateful to C. Boggs, A. Brower, M. Cardoso, I. Hanski, N. Janz, S. Nylin, J. Painter, C. D. Thomas, and an anonymous reviewer for comments on the manuscript. Discussions with E. Haukioja and M. C. Singer have helped me understand phytophagy both from the plants' view and the insects' view. I thank G. Beccaloni for information on host-plant use of South American species, S. R. Jensen for information on iridoids, L. McDade for information on the phylogeny of Acanthaceae, and R. Olmstead for information on the phylogeny of Asteridae. This study was funded by the Academy of Finland (grants 38604 and 44887 to I. Hanski).

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APPENDIX

Host plants of Melitaeini species. Sources: (1) Wahlberg, pers. obs.; (2) Denno and Benrey 1997; (3) DeVries 1987; (4) Feldman and Haber 1998; (5) Hesselbarth et al. 1995; (6) Kuussaari et al. 1995; (7) Luckens 1985; (8) Dyer and Floyd 1993; (9) Scott 1986, 1994, 1998; (10) Thomas et al. 1990; (11) Tolman 1997; (12) Wahlberg 1997; (13) Wahlberg 1998; (14) Weidemann 1988; (15) White and Singer 1974; Radtkey and Singer 1995; (16) Williams et al. 1984; (17) M. Singer, pers. comm. 1999; (18) G. Beccaloni, pers. comm. 1999; (19) S. van Nouhuys, pers. comm. 2000.

Species	Host plant family	Host plant species	Source	
<i>Euphydryas editha</i>	Orobanchaceae	<i>Pedicularis semibarbata</i>	15	
		<i>Pedicularis densiflorus</i>	15	
		<i>Castilleja nana</i>	15	
		<i>Castilleja breweri</i>	15	
		<i>Castilleja applegatei</i>	15	
		<i>Castilleja pilosa</i>	15	
		<i>Castilleja wightii</i>	15	
		<i>Castilleja foliolosa</i>	15	
		<i>Mimulus whitneyi</i>	15	
		Plantaginaceae	<i>Collinsia tinctoria</i>	15
			<i>Collinsia torreyi</i>	15
			<i>Collinsia parviflora</i>	15
			<i>Collinsia heterophylla</i>	15
			<i>Collinsia bartsiiifolia</i>	15
	<i>Collinsia sparsiflora</i>		15	
	<i>Orthocarpus densiflorus</i>		15	
	<i>Penstemon heterodoxus</i>		15	
	<i>Veronica serpyllifolia</i>		10	
	<i>Plantago lanceolata</i>		15	
	<i>Plantago erecta</i>		15	
	<i>Plantago insularis</i>		15	
	<i>Plantago hookeriana</i>		15	
	<i>Euphydryas phaeton</i>	Caprifoliaceae	<i>Lonicera interrupta</i>	9
		Plantaginaceae	<i>Chelone glabra</i>	9
			<i>Aureolaria flava</i>	9
			<i>Plantago lanceolata</i>	9
			<i>Penstemon hirsutus</i>	9
<i>Gerardia grandiflora</i>			9	
Orobanchaceae			<i>Pedicularis canadensis</i>	9
			<i>Seymeria macrophylla</i>	9
Caprifoliaceae			<i>Lonicera canadensis</i>	9
Lamiaceae			<i>Galeopsis tetrahit</i>	9
Oleaceae			<i>Fraxinus americana</i>	9
Valerianaceae			<i>Valerianella radiata</i>	9
<i>Euphydryas chalcedona</i>			Scrophulariaceae	<i>Scrophularia californica</i>
	<i>Diplacus auranticus</i>	10		
	<i>Buddleja davidii</i>	10		
	Plantaginaceae	<i>Penstemon newberryi</i>	10	
		<i>Penstemon breviflorus</i>	10	
		<i>Penstemon microphyllum</i>	10	
		<i>Besseyia plantaginea</i>	9	
		<i>Collinsia tinctoria</i>	10	
		<i>Collinsia sparsiflora</i>	17	
		Orobanchaceae	<i>Castilleja applegatei</i>	10
			<i>Castilleja affinis</i>	10
			<i>Castilleja chromosa</i>	10
			<i>Castilleja foliolosa</i>	10
	<i>Pedicularis semibarbata</i>		10	
	<i>Pedicularis densiflorus</i>		10	
	<i>Orobanche fasciculata</i>		10	
	<i>Trichostema lanatum</i>		10	
	Lamiaceae	<i>Symphoricarpos albus</i>	10	
	Caprifoliaceae	<i>Lonicera</i> sp.	10	
	<i>Euphydryas aurinia</i>	Dipsacaceae	<i>Succisa pratensis</i>	11
			<i>Scabiosa comosa</i>	1
<i>Scabiosa columbaria</i>			11	
<i>Cephalaria leucantha</i>			11	
<i>Knautia arvensis</i>			11	
Gentianaceae			<i>Gentiana alpina</i>	11
			<i>Gentiana kochiana</i>	11
Caprifoliaceae		<i>Lonicera etrusca</i>	11	
		<i>Lonicera implexa</i>	11	

APPENDIX. Continued.

Species	Host plant family	Host plant species	Source
<i>Euphydryas desfontainii</i>	Dipsacaceae	<i>Dipsacus fullonum</i>	11
		<i>Cephalaria leucantha</i>	11
		<i>Knautia arvensis</i>	11
<i>Euphydryas cynthia</i>	Plantaginaceae	<i>Plantago alpina</i>	11
<i>Euphydryas gillettii</i>	Caprifoliaceae	<i>Lonicera involucrata</i>	16
		<i>Symphoricarpos albus</i>	9
	Valerianaceae	<i>Valeriana occidentalis</i>	16
		Plantaginaceae	<i>Veronica wormskjoldii</i>
	Orobanchaceae	<i>Pedicularis bracteosa</i>	16
		<i>Castilleja miniata</i>	9
		<i>Castilleja linariaefolia</i>	16
<i>Euphydryas iduna</i>	Plantaginaceae	<i>Veronica alpina</i>	11
		<i>Veronica fruticans</i>	11
<i>Euphydryas intermedia</i>	Caprifoliaceae	<i>Lonicera caerulea</i>	7
<i>Euphydryas maturna</i>	Orobanchaceae	<i>Melampyrum pratense</i>	13
	Plantaginaceae	<i>Veronica longifolia</i>	13
	Oleaceae	<i>Fraxinus excelsior</i>	14
	Adoxaceae	<i>Viburnum opulus</i>	13
	Caprifoliaceae	<i>Lonicera xylosteum</i>	13
	Acanthaceae	<i>Siphonoglossa pilosella</i>	9
	Asteraceae	<i>Machaeranthera phyllocephala</i>	9
<i>Phyciodes vesta</i>	Convolvulaceae	<i>Convolvulus arvensis</i>	9
		<i>Cirsium andersonii</i>	9
<i>Phyciodes picta</i>	Asteraceae	<i>Cirsium vulgare</i>	9
<i>Phyciodes orseis</i>	Asteraceae	<i>Cirsium ochrocentrum</i>	9
<i>Phyciodes mylitta</i>	Asteraceae	<i>Cirsium neomexicanum</i>	9
<i>Phyciodes pallida</i>	Asteraceae	<i>Cirsium ochrocentrum</i>	9
<i>Phyciodes phaon</i>	Verbenaceae	<i>Lippia nodiflora</i>	9
		<i>Lippia lanceolata</i>	9
		<i>Justicia ovata</i>	9
<i>Phyciodes tharos</i>	Asteraceae	<i>Aster praealtus</i>	9
		<i>Aster texanus</i>	9
		<i>Aster lanceolatus</i>	9
		<i>Aster ericoides</i>	9
		<i>Aster laevis</i>	9
		<i>Aster laevis</i>	9
		<i>Aster simplex</i>	9
<i>Phyciodes cocyta</i>	Asteraceae	<i>Aster undulatus</i>	9
		<i>Aster laevis</i>	9
		<i>Aster glaucoides</i>	9
		<i>Aster hesperius</i>	9
		<i>Aster greatai</i>	9
		<i>Aster childensis</i>	9
		<i>Aster conspicuum</i>	9
		<i>Aster occidentalis</i>	9
		<i>Aster porteri</i>	9
		<i>Aster foliaceus</i>	9
<i>Tegosa anieta</i>	Asteraceae	<i>Mikania micrantha</i>	3
<i>Anthanassa ardys</i>	Acanthaceae	<i>Dicliptera unguiculata</i>	4
		<i>Pseuderanthemum cuspidatum</i>	4
		<i>Justicia valerii</i>	4
<i>Anthanassa texana</i>	Acanthaceae	<i>Justicia ovata</i>	9
		<i>Dicliptera brachiata</i>	9
		<i>Jacobinia carnea</i>	9
		<i>Ruellia carolinensis</i>	9
		<i>Pseuderanthemum cuspidatum</i>	4
<i>Anthanassa tulcis</i>	Acanthaceae	<i>Dicliptera unguiculata</i>	4
<i>Anthanassa ptolyca</i>	Acanthaceae	<i>Justicia</i> sp.	18
<i>Eresia eunice</i>	Acanthaceae	<i>Fittonia</i> sp.	18
<i>Eresia eranites</i>	Acanthaceae	<i>Odontonema</i> sp.	3
<i>Eresia myia</i>	Acanthaceae	<i>Justicia</i> sp.	3
<i>Texola elada</i>	Acanthaceae	<i>Justicia</i> sp.	3
		<i>Siphonoglossa pilosella</i>	9
<i>Dymasia dymas</i>	Asteraceae	unknown sp.	9
<i>Chlosyne gaudealis</i>	Acanthaceae	<i>Siphonoglossa pilosella</i>	9
<i>Chlosyne narva</i>	Amaranthaceae	<i>Justicia</i> sp.	3
		<i>Amaranthus</i> sp.	3
<i>Chlosyne janais</i>	Acanthaceae	<i>Odontonema</i> sp.	8
		<i>Anisacanthus wrightii</i>	2
		<i>Odontonema callistachyum</i>	2

APPENDIX. Continued.

Species	Host plant family	Host plant species	Source
<i>Chlosyne cyneas</i>	Orobanchaceae	<i>Seymeria tenuisecta</i>	9
<i>Chlosyne fulvia</i>	Orobanchaceae	<i>Castilleja integra</i>	9
		<i>Castilleja lanata</i>	9
<i>Chlosyne leanira</i>	Orobanchaceae	<i>Castilleja foliolosa</i>	9
		<i>Castilleja affinis</i>	9
		<i>Castilleja martinii</i>	9
<i>Chlosyne theona</i>	Orobanchaceae	<i>Castilleja lanata</i>	9
	Scrophulariaceae	<i>Leucophyllum texanum</i>	9
		<i>Leucophyllum frutescens</i>	9
		<i>Brachiostylisma</i> sp.	9
<i>Chlosyne nycteis</i>	Asteraceae	<i>Rudbeckia laciniata</i>	9
		<i>Actinomeris alternifolia</i>	9
		<i>Aster puniceus</i>	9
		<i>Aster umbellatus</i>	9
		<i>Helianthus divaricatus</i>	9
		<i>Helianthus tuberosus</i>	9
		<i>Helianthus strumosus</i>	9
		<i>Helianthus annuus</i>	9
		<i>Helianthus decapetalus</i>	9
		<i>Verbesina helianthoides</i>	9
		<i>Verbesina virginica</i>	9
		<i>Conyza canadensis</i>	9
<i>Chlosyne gorgone</i>	Asteraceae	<i>Helianthus annuus</i>	9
		<i>Helianthus pumilus</i>	9
		<i>Helianthus petiolaris</i>	9
		<i>Helianthus laetiflorus</i>	9
		<i>Helianthus trachelifolius</i>	9
		<i>Ambrosia trifida</i>	9
		<i>Iva xanthifolia</i>	9
		<i>Viguiera multiflora</i>	9
<i>Chlosyne californica</i>	Asteraceae	<i>Helianthus annuus</i>	9
		<i>Viguiera deltoidea</i>	9
<i>Chlosyne lacinia</i>	Asteraceae	<i>Helianthus annuus</i>	9
		<i>Calyptocarpus vialis</i>	9
		<i>Gaillardia pulchella</i>	9
		<i>Heterotheca latifolia</i>	9
		<i>Parthenium hysterophorus</i>	9
		<i>Silphium asperimum</i>	9
		<i>Simsia calva</i>	9
		<i>Viguiera dentata</i>	9
		<i>Xanthium pennsylvanicum</i>	9
		<i>Zexmenia hispida</i>	9
		<i>Ambrosia trifida</i>	9
		<i>Verbesina encelioides</i>	9
<i>Chlosyne harrisii</i>	Asteraceae	<i>Aster umbellatus</i>	9
<i>Chlosyne palla</i>	Asteraceae	<i>Aster radulinus</i>	9
		<i>Aster occidentalis</i>	9
		<i>Aster conspicuus</i>	9
		<i>Chrysothamnus nauseosus</i>	9
		<i>Erigeron speciosus</i>	9
		<i>Solidago californica</i>	9
		<i>Senecio triangularis</i>	9
<i>Chlosyne neumoegeni</i>	Asteraceae	<i>Acamptopappus shockleyi</i>	9
		<i>Acamptopappus sphaerocephalus</i>	9
		<i>Machaeranthera tortifolia</i>	9
<i>Chlosyne acastus</i>	Asteraceae	<i>Machaeranthera canescens</i>	9
		<i>Machaeranthera viscosa</i>	9
		<i>Chrysothamnus viscidiflorus</i>	9
<i>Poladryas arachne</i>	Plantaginaceae	<i>Penstemon cobaea</i>	9
		<i>Penstemon speciosus</i>	9
		<i>Penstemon dasyphyllus</i>	9
<i>Melitaea trivialis</i>	Scrophulariaceae	<i>Verbascum thapsus</i>	11
		<i>Verbascum densiflorum</i>	11
		<i>Verbascum longifolium</i>	11
		<i>Verbascum speciosum</i>	11
		<i>Verbascum delpicum</i>	11
<i>Melitaea punica</i>	Asteraceae	<i>Centaurea</i> sp.	5

APPENDIX. Continued.

Species	Host plant family	Host plant species	Source
<i>Melitaea scotosia</i>	Asteraceae	<i>Stemmacantha uniflora</i>	1
<i>Melitaea phoebe</i>	Asteraceae	<i>Centaurea scabiosa</i>	14
		<i>Centaurea montana</i>	11
		<i>Centaurea jacea</i>	11
		<i>Centaurea graeca</i>	11
		<i>Centaurea nigrescens</i>	11
		<i>Centaurea alba</i>	11
		<i>Centaurea columbaria</i>	11
		<i>Stemmacantha uniflora</i>	1
<i>Melitaea persea</i>	Plantaginaceae	<i>Linaria vulgaris</i>	11
<i>Melitaea deserticola</i>	Plantaginaceae	<i>Linaria aegyptiaca</i>	11
<i>Melitaea didyma</i>	Plantaginaceae	<i>Linaria vulgaris</i>	14
		<i>Plantago lanceolata</i>	14
		<i>Plantago media</i>	14
	Scrophulariaceae	<i>Verbascum lychnitis</i>	14
	Lamiaceae	<i>Stachys recta</i>	14
<i>Melitaea latonigena</i>	Plantaginaceae	<i>Veronica incana</i>	1
<i>Melitaea interrupta</i>	Plantaginaceae	<i>Plantago lanceolata</i>	5
<i>Melitaea didymoides</i>	Plantaginaceae	<i>Veronica incana</i>	1
<i>Melitaea arduinna</i>	Asteraceae	<i>Centaurea behen</i>	11
		<i>Centaurea nemecii</i>	11
<i>Melitaea cinxia</i>	Plantaginaceae	<i>Veronica spicata</i>	6
		<i>Veronica incana</i>	1
		<i>Veronica teucrium</i>	14
		<i>Plantago lanceolata</i>	11
		<i>Plantago maritima</i>	11
		<i>Plantago media</i>	6
		<i>Plantago alpina</i>	17
<i>Melitaea diamina</i>	Valerianaceae	<i>Valeriana sambucifolia</i>	12
		<i>Valeriana dioica</i>	14
		<i>Valeriana officinalis</i>	11
<i>Melitaea parthenoides</i>	Plantaginaceae	<i>Plantago lanceolata</i>	1
		<i>Plantago media</i>	14
		<i>Plantago alpina</i>	11
<i>Melitaea varia</i>	Gentianaceae	<i>Gentiana verna</i>	11
		<i>Gentiana acaulis</i>	11
	Plantaginaceae	<i>Plantago alpina</i>	11
<i>Melitaea aurelia</i>	Plantaginaceae	<i>Plantago lanceolata</i>	11
		<i>Plantago media</i>	19
<i>Melitaea deione</i>	Plantaginaceae	<i>Plantago lanceolata</i>	1
		<i>Linaria vulgaris</i>	11
		<i>Linaria alpina</i>	11
		<i>Linaria minor</i>	11
<i>Melitaea britomartis</i>	Plantaginaceae	<i>Veronica teucrium</i>	2
		<i>Plantago lanceolata</i>	11
<i>Melitaea athalia</i>	Plantaginaceae	<i>Veronica chamaedrys</i>	11
		<i>Veronica spicata</i>	1
		<i>Veronica montana</i>	11
		<i>Veronica officinalis</i>	11
		<i>Digitalis purpurea</i>	11
		<i>Plantago lanceolata</i>	14
		<i>Plantago alpina</i>	11
	Orobanchaceae	<i>Melampyrum pratense</i>	11
		<i>Melampyrum sylvaticum</i>	11
		<i>Pedicularis lapponica</i>	11