

# HOST PLANT USE AMONG CLOSELY RELATED *Anaea* BUTTERFLY SPECIES (LEPIDOPTERA, NYMPHALIDAE, CHARAXINAE)

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(With 1 figure)

## ABSTRACT

There is a great number of Charaxinae (Lepidoptera: Nymphalidae) species in the tropics whose larvae feed on several plant families. However the genus *Anaea* is almost always associated with *Croton* species (Euphorbiaceae). This work describes patterns of host plant use by immature and adult abundance on different vertical strata of sympatric *Anaea* species in a forest of Southeastern Brazil. Quantitative samples of leaves were taken in April/1999 and May/2000 to collect eggs and larvae of four *Anaea* species on *C. alchorneicarpus*, *C. floribundus* and *C. salutaris* in a semideciduous forest. Sampled leaves were divided into three classes of plant phenological stage: saplings, shrubs and trees. The results showed that the butterfly species are segregating in host plant use on two scales: host plant species and plant phenological stages. *C. alchorneicarpus* was used by only one *Anaea* species, whereas *C. floribundus* was used by three species and *C. salutaris* by four *Anaea* species. There was one *Anaea* species concentrated on sapling, another on sapling/shrub and two others on shrub/tree leaves. Adults of *Anaea* were more frequent at canopy traps but there were no differences among species caught in traps at different vertical positions. This work supplements early studies on host plant use among Charaxinae species and it describes how a guild of closely related butterfly species may be organized in a complex tropical habitat.

*Key words:* Nymphalidae, Charaxinae, *Anaea*, *Memphis*, *Croton*.

## RESUMO

### Uso de plantas hospedeiras entre espécies proximamente aparentadas de borboletas

Borboletas *Anaea* (Lepidoptera: Charaxinae) estão frequentemente associadas a espécies de *Croton* (Euphorbiaceae). Este trabalho descreve a utilização de plantas hospedeiras por uma guilda de borboletas em uma floresta do Sudeste brasileiro. Folhas de três espécies de *Croton* foram amostradas em 1999/2000 para a coleta de ovos e larvas de quatro espécies de *Anaea*. Paralelamente, foram colocadas armadilhas para adultos em duas diferentes posições verticais (1 m e 5 m) para comparação do número de indivíduos capturados entre as espécies. Os resultados demonstraram que a guilda de borboletas segrega na utilização de plantas hospedeiras em duas escalas: espécie hospedeira e estágio fenológico das plantas. *Croton alchorneicarpus* foi utilizada como hospedeira apenas por *A. ryphea*; *C. floribundus* foi utilizada por três espécies de *Anaea*; e *C. salutaris*, por quatro espécies. *Anaea otrere* foi mais abundante em plantas jovens, *A. ryphea* predominou em plantas jovens e arbustos e *A. appias* e *A. arginussa* apresentaram maiores densidades em folhas de arbustos e árvores de *Croton*. Os adultos de *Anaea* foram capturados com maior frequência nas armadilhas postas na copa das árvores, todavia não houve diferença significativa entre as espécies. Este trabalho

suplementa os dados sobre utilização de plantas hospedeiras por Charaxinae e descreve como uma guilda de borboletas proximamente aparentadas pode estar organizada em um ambiente tropical complexo.

*Palavras-chave:* Nymphalidae, Charaxinae, *Anaea*, *Memphis*, *Croton*.

## INTRODUCTION

Groups of closely related butterfly species are almost always confined to related larval host plants (Gilbert & Singer, 1975). There is a great number of Charaxinae (Lepidoptera: Nymphalidae) species in the tropics whose larvae feed on several plant families (DeVries, 1987). Ackery (1988) showed that many species feeding on Euphorbiaceae belong to the *Anaeini* tribe. The genus *Croton*, pantropical in distribution and having more than 800 described species (Webster, 1994), is the main larval foodplant and hosts several species from *Anaeini* in the genera *Hypna*, *Anaea*, *Polygrapha*, *Fountainea* and *Memphis* (Ackery, 1988).

The use of generic names in Charaxinae varies considerably among authors and I will follow Comstock's (1961) nomenclature, which considered *Memphis* Hübner as a subgenus of *Anaea* Hübner (see Caldas, 1994). The genus *Anaea* includes most Neotropical Charaxinae (e.g., DeVries, 1987, 1988). Biological studies for several *Anaea* species can be found in Muyschondt (1974, 1975a,b) and Caldas (1991, 1994). Up to twelve *Anaea* (*Memphis*) species have been recorded feeding on *Croton* (see Ackery, 1988), but in Brazil only *A. ryphea* Cramer has been studied more extensively (see Caldas, 1991, 1994, 1995).

Tropical adult butterflies may exhibit stratified distributions between canopy and understory (DeVries, 1988; DeVries *et al.*, 1997, 1999; Beccaloni, 1997). In some cases, larvae of butterfly species may be more abundant on saplings than on adults of the same host plant species (see Karban, 1987). Several others immatures of herbivore species have shown vertical stratification on their host plants (e.g., Brown *et al.*, 1997 and included references). In Ithomiinae the flight height of adults may be correlated to the occurrence of their larval host-plants (Beccaloni, 1997). Charaxinae species have often been trapped in the

forest canopy (DeVries, 1988; DeVries *et al.* 1997; DeVries *et al.* 1999) but stratification of immatures was not investigated. This work describes patterns of host plant use by immatures of sympatric *Anaea* species that feed on *Croton* and compares the abundance of adults between understory and canopy strata in a forest of Southeastern Brazil.

## METHODS

Three *Croton* species are commonly found along trails in the semideciduous forest of the Serra do Japi Reserve (23°16'S, 47°00'W) near Jundiá, São Paulo, Brazil, immediately after Japi. The place is a continuous forest of 300 km<sup>2</sup> with an average canopy height ranging between 5 and 15 m above the ground. *Croton floribundus* Spreng is the most widespread and abundant species and *C. salutaris* Baill is the rarest species and occurs in discrete patches. A third species, *C. alchorneicarpus* Croizat, may be very abundant on some trails but is rare on others.

Eggs of *Anaea* species (1.0 mm diameter) are frequently laid singly on the underside of leaves. Although they are very similar in appearance, there are some variations of color among species. Larvae from the first and second instar are quite similar to the naked eye, but from the third instar onward different species can be easily identified in the field. First to third instar larvae of *Anaea* species construct frass chains, adding feces with silk to the leaf vein, on the tip of which they rest. Fourth and fifth instar larvae may roll the leaf to form a tubular shelter, holding it closed with silk, and hide inside when not feeding (see Muyschondt, 1974, 1975a,b, and Caldas, 1994, for descriptions of immatures).

I used a stratified sample of leaves in April/1999 and May/2000 to collect eggs and larvae of *Anaea* on the three *Croton* species in Japi. I took the samples at four different sites in the forest. These sites were as far as 4 km from each other.

In 1999 I sampled leaves up to 3 m height and in 2000 I divided the samples into three classes: leaves sampled on saplings (less than 1m height), on shrubs (from 1-3 m) and on trees (from 3-5 m). Eggs and first instar larvae obtained in quantitative samples were reared in the laboratory using closed plastic dishes containing foodplant leaves and identified to species after rearing to the third instar. Larvae species from the third instar onward were identified in the field. At the end of June/2000 I used paired traps with rotting banana bait (see trap design in DeVries, 1987) to sample adults of *Anaea* at two heights. One trap of each pair was placed at 5 m from forest floor in the forest canopy and the other immediately below it, 1m from ground level. I positioned traps to receive direct sunlight between 10:00 to 14:00 h (the sample time). All butterflies caught in traps after the 4 hour period were identified and set free. Total sample effort was 92 hours of trapping for each height in a total of seven days from two to four different sites in the forest.

## RESULTS

In all, over 16831 *Croton* leaves were inspected in two years of sampling. Leaves from *C. floribundus* accounted for 55%; from *C. alchorneicarpus* for 25% and from *C. salutaris* for 20% of samples. The quantitative sample showed that *C. alchorneicarpus*

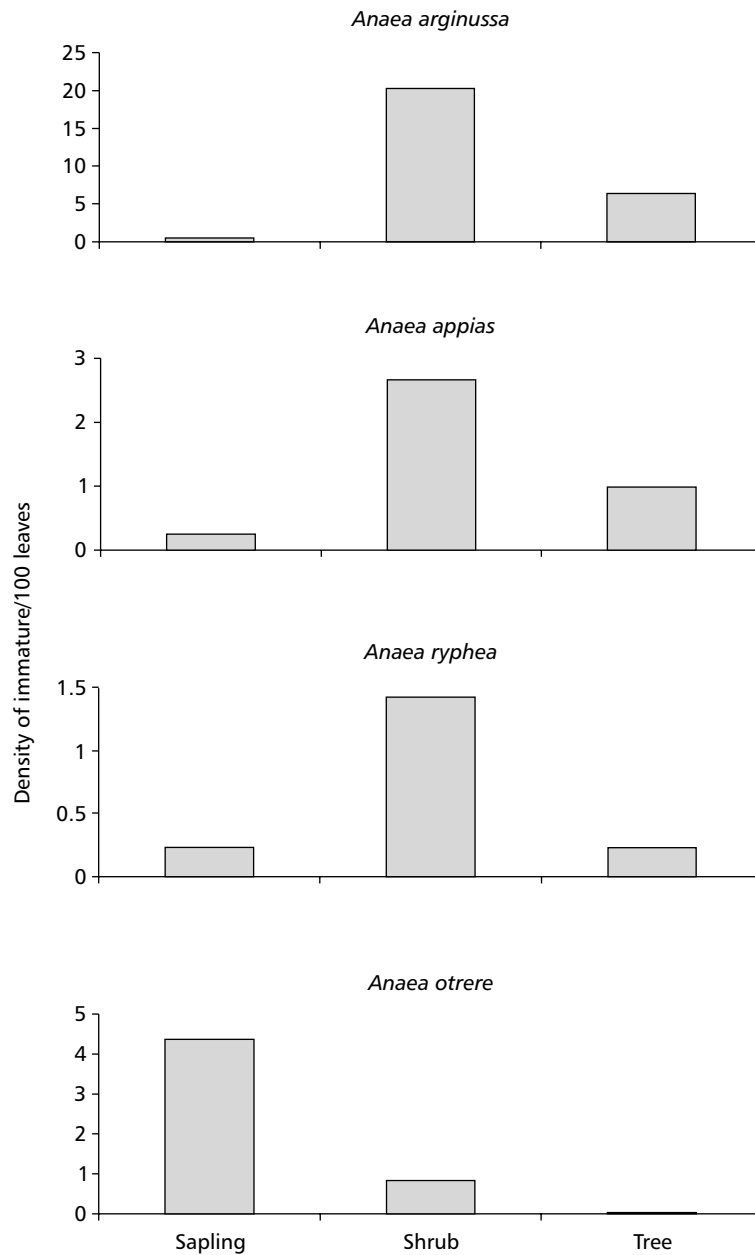
was used only by *A. ryphea* in Japi. Indeed I have never seen other *Anaea* species using *C. alchorneicarpus* but another Charaxinae, *Hypna clytemnestra* Butler, was sporadically recorded. *Croton floribundus* was used by all *Anaea* species except *A. arginussa* (Geyer). *Croton salutaris* hosts *A. otrere* (Hübner), *A. appias* (Hübner), *A. ryphea* and mainly *A. arginussa*. *Anaea otrere* and *A. appias* were predominantly found on *C. floribundus*. *Anaea arginussa* only occurred on leaves of *C. salutaris* and *A. ryphea* was most frequently seen in *C. alchorneicarpus* leaves (Table 1).

Densities of immatures *A. appias* and *A. otrere* were very similar in 2000. Although both species use *C. floribundus* as their main larval foodplant, *A. otrere* was predominantly found on saplings whereas *A. appias* was concentrated on leaves of shrubs and, in a lesser degree, in trees. Moreover, *Anaea otrere* was also abundant on sapling leaves of *C. salutaris*. On the other hand, *A. arginussa*, that shared *C. salutaris* leaves with *A. otrere*, was predominantly found on shrub and tree leaves. *Anaea ryphea* was more abundant in shrubs, mainly *C. alchorneicarpus*. Although *A. ryphea* occurred on leaves of trees, the densities on those leaves in relation to shrub leaves were much lower in comparison with *A. appias* and *A. arginussa*. Therefore *A. appias* and *A. arginussa* occurred more frequently than *A. ryphea* in trees (Fig. 1).

TABLE 1

Frequency of immature *Anaea* species occurrence on different *Croton* species in the Serra do Japi Reserve, Brazil. Data for eggs and larvae were lumped together. In parentheses, total number of sampled leaves and data in percentage. In 1999 immatures of *A. appias* were absent from sampled leaves.

	<i>C. floribundus</i>		<i>C. alchorneicarpus</i>		<i>C. salutaris</i>	
	1999 (3225)	2000 (6087)	1999 (1134)	2000 (3142)	1999 (564)	2000 (2679)
<i>A. otrere</i>	75 (94%)	81 (74%)	0 (0%)	0 (0%)	5 (6%)	28 (26%)
<i>A. appias</i>	0	104 (93%)	0	0 (0%)	0	8 (7%)
<i>A. ryphea</i>	6 (14%)	20 (28%)	36 (82%)	51 (72%)	2 (4%)	0 (0%)
<i>A. arginussa</i>	0 (0%)	0 (0%)	0 (0%)	0 (0%)	60 (100%)	202 (100%)



**Fig. 1** — Densities of immatures *Anaea* species among plant phenological stages. Data for eggs and larvae were lumped together. Densities for *A. otrere* and *A. appias* were calculated considering the number of sampled leaves of *C. floribundus* and *C. salutaris*. Densities for *A. arginussa* were calculated considering only sampled leaves of *C. salutaris* and densities for *A. ryphea* were calculated considering sampled leaves of all *Croton* species. These calculations were based on natural occurrence data of *Anaea* species among host plant species in 1999 and 2000 samples (see Table 1). Number of sampled leaves for each phenological stage in 2000 were: Saplings (2297), Shrubs (5546) and Trees (4065).

TABLE 2

Number of individuals of *Anaea* species and position of paired traps. Total of baiting was 92 hours in seven days of sampling for each height (between 10:00-14:00 h). There were no significant statistical differences among *Anaea* species in the number of individuals captured by the two vertical trap positions (G test;  $G = 5.9286$ ;  $p > 0.05$ , d.f. = 3).

<i>Anaea</i> species	Trap position		Total
	1 m	5 m	
<i>A. appias</i>	24	106	130
<i>A. arginussa</i>	3	13	16
<i>A. otrere</i>	2	2	4
<i>A. ryphea</i>	13	24	37
Total	42	145	187

These results suggest that a butterfly guild composed of closely related species is segregating in host plant use at least on two scales: host plant species and plant phenological stages (see Table 1 and Fig. 1).

The number of *Anaea* adults trapped in seven days was 187; there were 3.5 times more adults captured at the 5 m positions than at the 1 m traps. *Anaea arginussa* and *A. appias* were trapped 4.5 more times in 5 m than at 1 m traps, whereas *A. ryphea* was only 2 times more frequent at canopy traps. *Anaea otrere* was equally frequent in either vertical strata but only 4 adults were trapped during the sampling. Even with the differences cited above, the number of captured individuals among *Anaea* species was not significantly different between understory and canopy traps (Table 2).

## DISCUSSION

Although Charaxinae butterflies use several host plant families (e.g., DeVries, 1987; Ackery, 1988), *Anaea* butterflies are frequently associated with *Croton* species (e.g., Ackery, 1988). The data reported here suggest that these butterflies are segregating in host plant use in two scales in Serra do Japi Reserve. The guild of *Anaea* butterflies occurred at different degrees of densities among *Croton* species and phenological stages. Females butterfly usually lay

their eggs on habitats where juvenile growth and survival are best (Rausher, 1979; Freitas & Oliveira, 1996). The co-occurrence of butterfly species on the same foodplant may affect the number of eggs laid and larval survivorship (Yamamoto, 1981). Thus if competition among *Anaea* species is important in structuring the community on *Croton*, then segregated use of larval foodplant may be advantageous.

In Santa Genebra Reserve (hereafter St. Genebra), a forest fragment in Campinas, SP, 50 km far from Japi, only *A. ryphea* had been recorded using *Croton* (Caldas, 1991). In this fragment *A. ryphea* is found on *C. floribundus* and *C. priscus* Croizat saplings and shrubs (Caldas, 1994). Although adults of *A. arginussa*, *A. appias* and *A. otrere* have been recorded in the area (K.S. Brown, pers. com.) caterpillars were rarely seen on *Croton* leaves (*A. Gomes-Filho* and myself, pers. obs.). Caldas (1991) recorded only one other Charaxinae, *Hypna clytemnestra*, using *Croton* in St. Genebra. She suggested that *H. clytemnestra* could be considered a potential competitor when it co-occurs with *A. ryphea*, however observations on asymmetric competition between these two species indicated a low-intensity interaction, because the *H. clytemnestra* population was never higher than 10% of the *A. ryphea* population (Caldas, 1991). However, I suggest that in Japi *A. otrere* is a strong candidate to be a competitor of *A. ryphea* because it frequently occurs in a high density on *C. floribundus* leaves.

The community of immature *Anaea* species on *Croton* leaves in Japi is much more diverse than in St. Genebra forest fragment. While in Sta. Genebra *C. floribundus* leaves are dominated by *A. ryphea* (Caldas, 1991), it seems that *A. ryphea* is displaced to use *C. alchorneicarpus* as its main larval foodplant in Japi. *Croton floribundus* saplings were the main foodplant for *A. otrere*, but leaves were frequently used by two other species in Japi. I suggest that *A. otrere* could be considered a specialist in saplings, due to the observed high densities on the two host plant species at those phenological stages. Unfortunately, the number of adults of *A. otrere* captured in the traps was too low to verify if adults were also stratifying vertically. In the field, I saw adults of *A. otrere* flying very close to the ground level apparently in oviposition behavior. An additional sampling effort should be conducted in the future to collect more adults of *A. otrere* in different vertical positions.

*Anaea appias* and *A. arginussa* that shared the same distribution pattern among plant phenological stages segregated in use of host species. These two species may be considered in a continuous pattern of host plant use as canopy species. Indeed the adults from these species were twice as frequent as *A. ryphea* in canopy traps. Moreover and interestingly they have a particular way to make a leaf funnel, different from the two other *Anaea* species. The funnel position of *A. appias* and *A. arginussa* is perpendicular to the leaf plane whereas in the other two species the funnel is at the same level of leaves. I suggest that the particular behavior of rolling leaves may be used as one of the taxonomic characters to separate *Anaea* species. It remains to be investigated if these different behaviors of making leaf refuges have any ecological role.

This study supplements Muysshondt's early works (1974, 1975a,b) and more recently Caldas' (1991, 1992, 1994) towards a better understanding of host use in Charaxinae and encourages other investigations of *Anaea* community structure in places like the Amazon Basin and Central America where there is a great diversity of these butterflies and *Croton* species. As DeVries *et al.* (1999) points out, the documentation

of diverse insect communities in space (as done here) and time (as done there) can reveal ecological patterns relevant to elucidate the evolution of community structure in complex tropical habitats. The system presented here deserves more attention. Future experimental studies with potted plants should be conducted to separate the effects of leaf quality among host plant phenological stages and the vertical positions of leaves per se, as well as tests of oviposition and host preference among *Anaea* species to help in explaining the observed segregation patterns.

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