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Cover Page Footnote

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Life History and Laboratory Rearing of the Red Admiral, *Vanessa atalanta* (Lepidoptera: Nymphalidae)

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Abstract

The red admiral butterfly, *Vanessa atalanta* (Linneaus, 1758) (Lepidoptera: Nymphalidae) is a globally distributed species and model organism for studying migration patterns and effects of climate change. Most previous red admiral research focused on wild populations. Establishing laboratory colonies allow for experimentation with a multitude of lab-based plant-insect interactions. We describe red admiral butterfly life history and laboratory rearing methods.

Keywords: Rearing techniques, Laboratory colony

The red admiral butterfly, *Vanessa atalanta* (Linneaus, 1758) (Lepidoptera: Nymphalidae), is a globally distributed species and model organism for studying mate-induced territorial defense, migration patterns, and effects of climate change (Field 1971, Brown and Alcock 1990, Benvenuti et al. 1994, Tolman and Lewington 1997, Roy and Sparks 2001, Mikkola 2003, Stefanescu 2008, Fox and Dennis 2010). In North America, red admirals are widely distributed from Guatemala to northern Canada. Within the United States, adults overwinter in southern Texas and southern Florida, migrating northwards during the spring in March through May (Opler 1992, Swanson and Monge-Nájera 2000). In southern states, they have four generations per year but are bivoltine in the northern regions (Scott 1986, Swanson and Monge-Nájera 2000). Red admirals have five larval instars with larvae folding leaves to form shelters on host plants (Simon et al. 1997, Hall and Butler 2021). Larvae consume plants within the family Urticaceae, such as stinging nettle (*Urtica dioica* L.), slender nettle (*Urtica dioica gracilis* [Aiton] Selander), wood nettle (*Laportea canadensis* [L.] Wedd.), false nettle (*Boehmeria cylindrica* [L.] Swartz), and in Cannabaceae, such as hops (*Humulus* spp.) (Evans 2008).

Previous sources provide guidelines for rearing butterflies including red admirals (Stout 2021). This manuscript focuses on maximizing colony production in laboratory settings. Most previous research has focused on wild populations of red admirals. Establishing laboratory colonies allows for experimentation with a multitude of lab-based plant-insect interactions and other basic research of these organisms. Here, we describe novel laboratory rearing methods for red admiral butterflies.

Materials and Methods

To create substrates for oviposition and provide food for larvae, we planted stinging nettle seeds (Sheffield's Seed Company, Locke, NY) in 4 × 6 × 6.5-cm trays with topsoil (Scott's Lawn Premium) and watered every 48 hrs. When plants had four true leaves, we transferred them into 2.84 L pots placed into sub-irrigation trays continuously filled with water, in an environmental chamber (28 °C, 16L : 8D).

During April 2019, we collected red admiral larvae (instars 1–5) feeding on slender nettle (*U. dioica gracilis*) from University of Missouri's Baskett Wildlife Research Center in Calloway County, Missouri. Larvae were transported to the laboratory and placed in individual petri dishes (95 × 15-mm polystyrene, Fisher Scientific) lined with filter paper and Plaster of Paris (Fig. 1). Each dish was soaked with deionized water, drained of excess water, then transferred to growth chambers (28 °C, 16L : 8D). We provided two stinging nettle leaves, removed frass, and replaced moistened filter paper every 48 hrs. After larvae pupated on leaves in the petri dishes, we transferred leaves with attached pupae to mesh cages (30 × 30 × 60-cm Bugdorm-2120) located in environmental chambers (28 °C, 16L : 8D), carefully maintaining pupae in the original horizontal position to avoid disrupting development.

Figure 1. Fourth instar red admiral in rearing dish with stinging nettle leaves.

Figure 2. Red admiral adult feeding on honey solution in adult rearing cage.

We provided emerging adults 25% honey and deionized water solution in a 3 × 6-cm plastic dish located on an 18-cm platform every 72 hrs (Fig. 2). Two potted stinging nettles, approx. 20 cm tall, were placed into each cage as an oviposition substrate (Fig. 3). Emerged adults mated in cages, and females laid individual eggs on potted stinging nettle foliage. Newly-hatched larvae were transferred from cages using paint brushes into individual rearing dishes, as described above. Life history data was recorded from egg to adult for the fully laboratory reared generation of 25 individuals feeding only on stinging nettle (*U. dioica*).

Figure 3. Adult rearing cages with feeding platform and stinging nettle plants for oviposition.

Results

Eggs hatched after five to seven days ($\bar{x}$ = 5.72, n = 25). First and second instars formed leaf shelters in petri dishes. Larvae feeding on stinging nettle foliage required an average of 18.3 days to reach pupation (n = 25), typically consuming one leaf per day during development. Adults emerged after an average of 8.7 days after pupation (n = 17). On average, larva required 27.83 days to develop to adult, with 68% of larvae surviving from egg to adult (n = 17).

Discussion

Using our methods, large numbers of red admirals can be produced with an acceptable survival rate (Shelton et al. 1991, Greenberg et al. 2001). With an average development time from egg to adult of 27.83 days (n = 17) and newly emerged females mating and ovipositing within a few days, the entire life cycle occurs within only approximately 30 days. Additionally, females laid multiple eggs on caged plants in the laboratory, whereas wild populations lay individual eggs on plants.

This method employs easily obtained supplies that improves efficiency of colony rearing red admiral butterflies in laboratories. Our method rears larvae in stacked petri dishes within growth chambers, and then transfers pupae to cages for adult mating and oviposition, as opposed to placing one larva per caged potted plant, thereby substantially reducing space required to maintain a laboratory colony. We provide leaf foliage to larvae in petri dishes, further reducing the amount of potted host plants required to maintain a colony. Late instar red admiral larvae will occasionally chew through the main stem to create leaf shelters, thus reducing the viability of the food

source when given access to the whole plant (Scott 1986).

Further experimentation might improve the survival rate of the red admirals reared under laboratory conditions. Of the 25 reared larvae, eight individuals failed to eclose from pupae. This might indicate that forming pupae in the petri dish or transferring pupae to the adult cage causes increased mortality. Alternative methods for maintaining pupae should be evaluated.

Stinging nettle are fast growing host plants and seeds are commercially available. However, in this study, wild caterpillars were collected on wild slender nettle, and then reared to adult on stinging nettle for supplying eggs to our colony. Switching host plant species might affect the survival rate of larvae and fecundity of adults (Stoyenoff et al. 1994, Greenberg et al. 2001, Saeed et al. 2010, Roy and Barik 2012) and might change with subsequent generations feeding only on stinging nettle (Hoffman and Ross 2018). Additionally, alternate plant hosts, such as slender nettle and false nettle, should be examined in future rearing trials specifically focusing on larvae development time, survival, and consumption rates. Host plants should also be assessed for maximum foliaged produced and quality per plant for laboratory rearing.

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